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ELECTRON MICROSCOPICAL STUDIES OF ULTRA-THIN SECTIONS IN *ASPERGILLUS, PENICILLIUM* AND *NEUROSPORA**

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(Accepted for publication September 14, 1955)

In the course of cytological studies in a few selected fungi by means of ultra-thin sectioning and electron microscopy, some new and significant information has been obtained concerning the conidia and mycelia of *Aspergillus oryzae*, *Penicillium chrysogenum* and *Neurospora crassa*.

The cytological study of fully developed fungi is rendered difficult by their extreme refractility. Acid hydrolysis makes the conidia stainable and has been valued as an effective means of demonstrating their nuclear material. Some cytological features of the conidia of *Aspergillus candidus* have already been described by the author (1951). The cytology of microorganisms involves a number of structures and events which are beyond the resolution limit of the microscope. In order to make them accessible to investigation, the materials have been cut into thin slices by means of the sectioning technique recently developed for electron microscopy. The study of these sections has clarified the normal organization of the cytological materials in the conidia and mycelia of fungi.

Cytological studies of microorganisms by means of electron microscope have been carried out by many authors, for instance, Hillier, Mudd, and Smith, (1949); Mudd, and Smith, (1950); Mudd, et al. (1950) (1951); Tsujita, Watanabe, and Tsuda, (1954); Sjostrand, (1954); Tsuda et al. (1954), and others.

MATERIALS AND METHODS

Conidia and mycelia of *Aspergillus oryzae*, *Penicillium chrysogenum* and *Neurospora crassa* have been examined. Some results of this study have already been described (Tsuda, 1951, 1953).

Aspergillus oryzae and *Neurospora crassa* were grown on wort agar medium at 28°C. The conidia and mycelia of these organisms were collected after 3 days and then the materials were washed repeatedly with distilled water before being prepared for sectioning. *Penicillium chrysogenum* was cultivated in submerged condition at 28°C. for 3 days, then the mycelium was collected by moderate centrifugation lasting ten minutes and washed in the same way as *Aspergillus* and *Neurospora*. To 1 ml. (approximately) suspension of the obtained material, 10 ml. of 0.5 per cent osmium tetroxide in phosphate buffered solution was added. The materials were stoppered and put aside for 30 minutes. The fixed conidia and mycelia

*Contributions from the National Institute of Genetics, Japan, No. 117.

were washed several times with distilled water, then dehydrated for one hour each in 70 per cent, 95 per cent, and 100 per cent ethanol and left one hour in each of three changes of absolute alcohol. From alcohol the materials were transferred, by alternate centrifugation, decanting, and resuspending, to a mixture of equal parts of n-butyl methacrylate and absolute alcohol (1 hour), then to pure monomeric n-butyl methacrylate (1 hour), and finally to a mixture of 95 parts of n-butyl methacrylate and 5 parts of methyl methacrylate with 2 per cent benzoyl peroxide as catalyst. At this stage a few drops of the suspension of the material were placed in No. 3 rectal gelation capsules and centrifuged for a few minutes at low speed. The preparations were kept at 47°C until polymerization was completed, which was usually accomplished within 24 hours. Before sectioning, the gelatin capsules were soaked off in water. Sections of less than 0.1μ were cut by Spencer's ultra-microtome. For collection the sections were floated off the knife edge on a surface of 40 per cent ethyl alcohol in distilled water. They were picked up on the electron microscope copper grids previously coated with an extremely thin film of formvar and dried at room temperature. Then the sections were carried through amyl acetate which removed the polymer. The specimens were examined under a J. E. M. type III electron microscope made by Japan Electron Optics Laboratory.

Results and Discussion

The *Aspergilli* are, as is well known, the most popular material for physiological studies of fungi, and though we have a considerable number of valuable reports in this field, yet it is rather strange to realize that our knowledge of the cytology of this group is very meagre. With regard to the nucleus of the *Aspergillaceae*, Strasburger (1902) recorded for the first time some observations on *Penicillium* species. Later, also Wakayama (1931) in his excellent work, described the nuclear division in the sterigma of *Aspergillus* species.

The *Aspergillaceae* are well characterized as a rule by the formation of a perithecium in their life cycle. But in many species, the development of the perithecium in nature is of rare occurrence and multiplication takes place chiefly by conidia.

The fungus *Neurospora crassa* is an ascomycete with an imperfect stage known as Monilia. The vegetative stage consists of branched, septate hyphae, each cell containing several nuclei. Reproduction of the vegetative phase occurs asexually by means of conidia which are multinucleate, by microconidia which are uninucleate, or by hyphal fragments. In my experiment, only the vegetative phase was examined.

Comprehensive cytological studies of the conidia of *Aspergillus* have been carried out by Ishitani and Sakaguchi who used the light microscope. The present author has also described some cytological features of those conidia. Fig. 1 shows that they have in most cases two nuclei. Only when heterocarya were formed, he could observe conidia with five or more nuclei (Tsuda, S. 1951). He also found that the conidia are hydrolyzed when placed for 7 minutes in N HCl

at 60°C. They could be stained hereafter with Giemsa, diluted by adding 1 drop of the stain to 1 cc. dist. water, and applied for about 15 minutes.

Fig. 2 is an electron micrograph of an ultra-thin section of *Aspergillus oryzae* showing a conidium with three nuclei ; the nuclear membrane is not clearly defined. The cell walls of the conidia are thick and rugged, as Fig. 3 (electron micrograph) shows ; they seem to be somewhat separated from the cytoplasm. The conidium to the right shows secondary nuclear division. The cytoplasm as well as the nuclei have a loose filamentous structure.

Figs. 4-9 illustrate the finer inner structure of the mycelia in various successive stages. The cell wall is thick and the cytoplasm has a fine reticulated appearance. Many granules occur among the cytoplasmic inclusions.

Figs. 4 and 5 are electron micrographs of ultra-thin longitudinal sections of hyphal cells of *Aspergillus oryzae.* Fig. 4 shows a mycelium fixed at a late stage during the formation of a transverse cell wall at the left end. A large amount of cytoplasmic granules is scattered in the reticulated cytoplasm (Fig. 5), among them very dense, spherical bodies of various size. In still later stages many vacuoles of various sizes could be observed, in electron micrograph obtained from an ultra-thin transverse and oblique section of bundled hyphae of an old culture (Fig. 6). The vacuoles increase in number and volume with the age of the cells. At last, one large vacuole occupies the wholl cell lumen (Fig. 6, right).

Figure 7 is an electron micrograph of an ultra-thin longitudinal section of a hyphal cell of *Neurospora crassa* shadowed with chromium. The cytoplasm shows a reticulated structure and many cytoplasmic granules as in *Aspergillus oryzae.* This figure shows the formation of a transverse cell wall (at the left) and hyphal cell nuclei (at the right). The nuclear material seems to be more or less reticulate, almost indistinguishable in density and structure from the cytoplasm. When seen through a light microscope, the properly stained structures could be easily interpreted as achromatic and chromatic figures. In the middle of the nucleus, a spherical body can be seen which corresponds to the nucleolus of higher plants. This spherical body has a very dense consistency. Unfortunately, recognition of individual chromosomes in fungi by means of electron microscope was not yet possible. At present no such structures have been observed which could be interpreted as the mitotic figures described by Wakayama (1931, 1932) and other authors.

The other structures of interest in the hyphal cells of fungi are cytoplasmic granules. Figures 8 and 9 are electron micrographs of ultra-thin connected oblique sections of the same cell of *Penicillium chrysogenum.* The cytoplasm shows a reticulated structure and many cytoplasmic granules as in *Aspergillus* and *Neurospora.* In *Penicillium chrysogenum,* the cross sections of the cytoplasmic granules are mostly spherical or elliptical, and are surrounded by a limiting membrane as can be seen from Figs. 8 and 9. The inner surface of

the inner layer of the limiting membrane exhibits parallel lamellae which seem to protrude into the interior of the cytoplasmic granules. Also the "cristae mitochondriales" so-called by Palade could be observed.

Studies on mitochondria-like granules in bacteria and fungi have been carried out by many authors. Mudd, et al. (1951) have examined certain bacteria by means of electron microscope, and Hartman, and Charlotte, (1954) have also studied *Saccharomyces cerevisiae* using the following staining technique. They have found that two indicators of cytochrome c plus cytochrome oxidase activity give comparable staining patterns in the wild type yeast cell. The first, the Nadi reagent, is oxidized to an insoluble product. The second indicator, Janus green B, possibly has two properties responsible for its specific staining of mitochondria, of which an inherent affinity for mitochondria may be one. Haskins, et al. (1953) and others have carried out biochemical studies on cytochromes and the oxidase system of *Neurospora*.

It may be argued that the present micrographs represent many artifacts. While this may be partially true, the preparations examined are certainly better than those obtained in the early electron microscope work in that they are well fixed and, since they have been subjected to a dehydration series and have been polymerized in plastic, they have not been subjected to the violent forces of surface tension while drying or of osmotic effects which arise during the washing of unfixed materials (Anderson. 1952). It now appears that what could be seen of the finer inner structure of conidia and mycelia was really beyond the limit of resolution of the light microscope. The study of the nuclear apparatus of fungi and, especially, cytoplasmic granules will be continued (Tsuda, 1956).

Acknowledgment

The writer wishes to express his hearty thanks to Dr. M. Tsujita and Mr. K. Gotoh for valuable suggestions and criticisms.

Summary

Electron microscopical studies of the inner structure of *Aspergillus oryzae*, *Penicillium chrysogenum* and *Neurospora crassa* have been carried out by means of ultra-thin sections. The results of the observations are summarized as follows.

The cell wall of the conidia of *Aspergillus oryzae* is thick and rugged, and it has the tendency to become somewhat separated from the cytoplasm. The conidia have several nuclei which have a nuclear membrane.

The hyphal cells of the examined fungi have thick cell walls and the cytoplasm shows a reticulated structure. A great amount of cytoplasmic granules are scattered in the reticulated cytoplasm. Among the cytoplasmic inclusions occur spherical bodies of various size.

In hyphal cells of *Penicillium* the author fortunately could observe the finer inner structure of the cytoplasmic granules which are surrounded by limiting membrane. On the inner surface of the inner layer of the limiting membrane lamellae and "cristae mitochondriales", structures found by Palade in cells of higher animals could be clearly observed.

Other bodies of interest in the hyphal cells of the fungi are the nuclei. The nuclear materials seem to have a more or less reticular structure, almost indistinguishable in density from the cytoplasm.

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Explanation of Figures

Fig. 1. Light micrograph of the conidia of *Aspergillus oryzae* stained with Giemsa solution after hydrolysis.

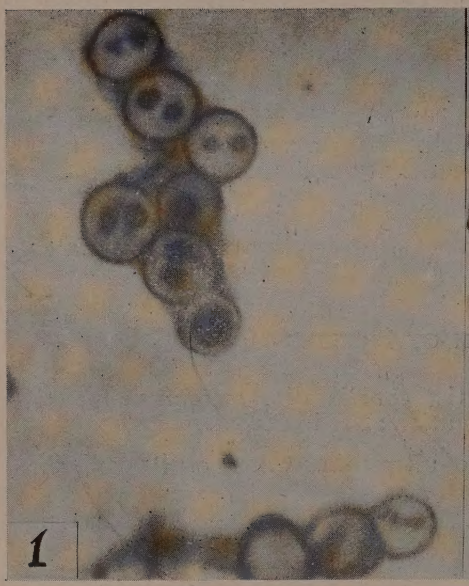
Figs. 2-3. Electron micrographs of ultra-thin sections of the conidia of *Aspergillus oryzae.*

Figs. 4-5. Electron micrographs of ultra-thin longitudinal sections of hyphal cells of *Aspergillus oryzae.*

Fig. 6. Electron micrograph of an ultra-thin transverse and oblique section of bundled hyphae from an old culture of *Aspergillus oryzae.* (shadowed with chromium).

Fig. 7. Electron micrograph of an ultra-thin longitudinal section of a hypha of *Neurospora crassa.* (shadowed with chromium).

Figs. 8-9. Electron micrographs of two ultra-thin oblique connected sections of a cell of *Penicillium chrysogenum.*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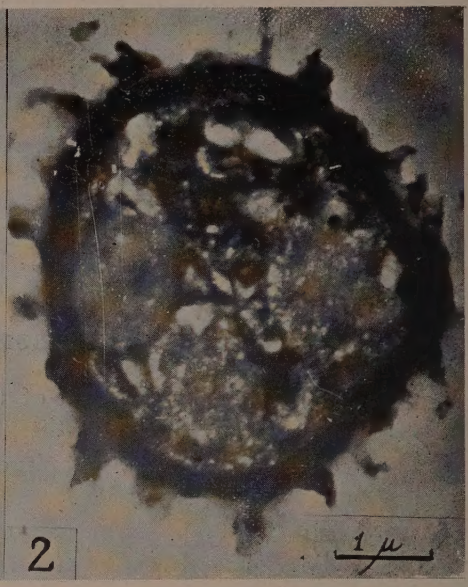
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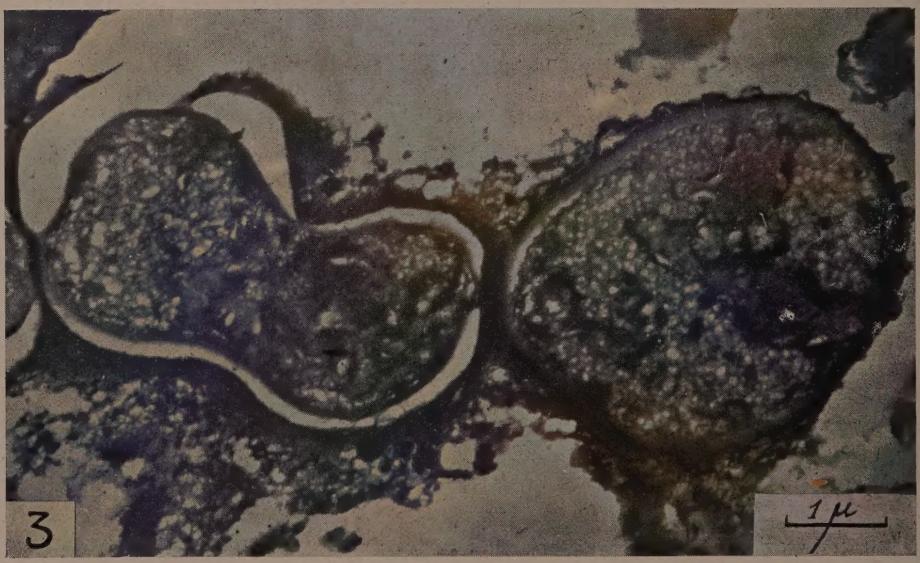
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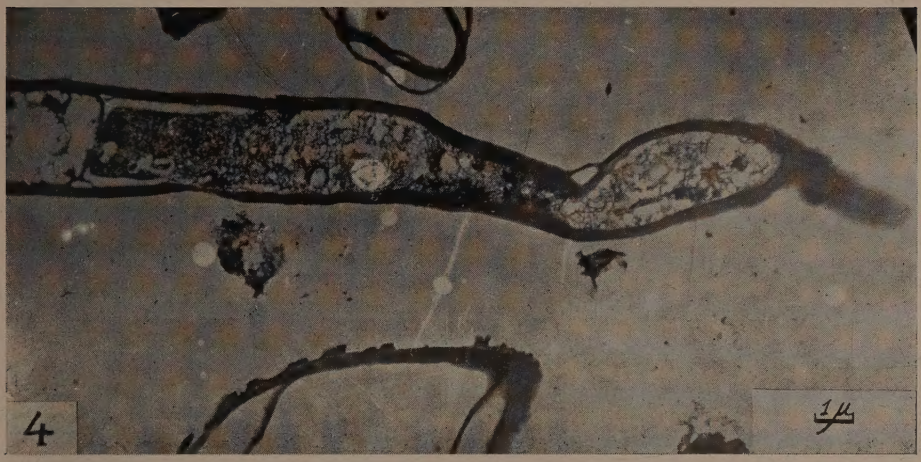
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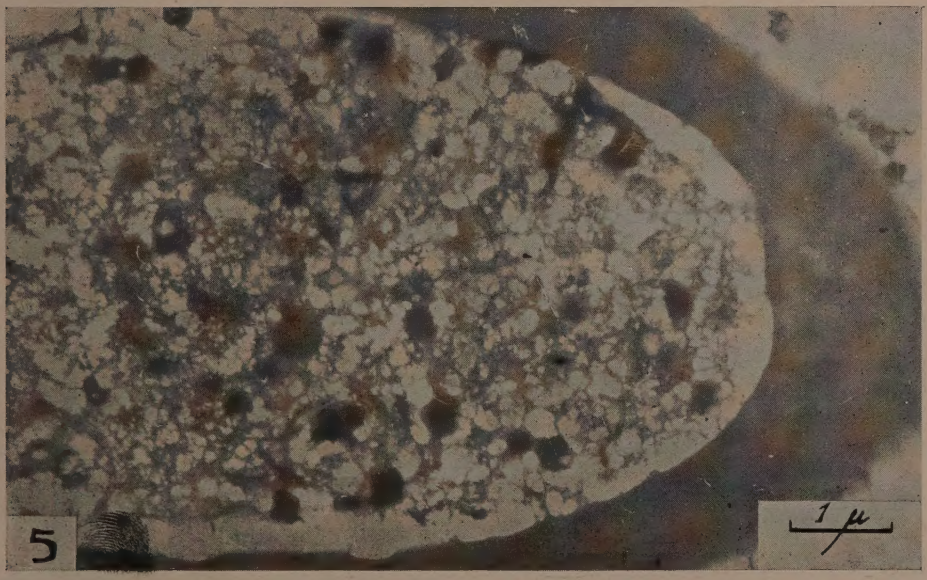
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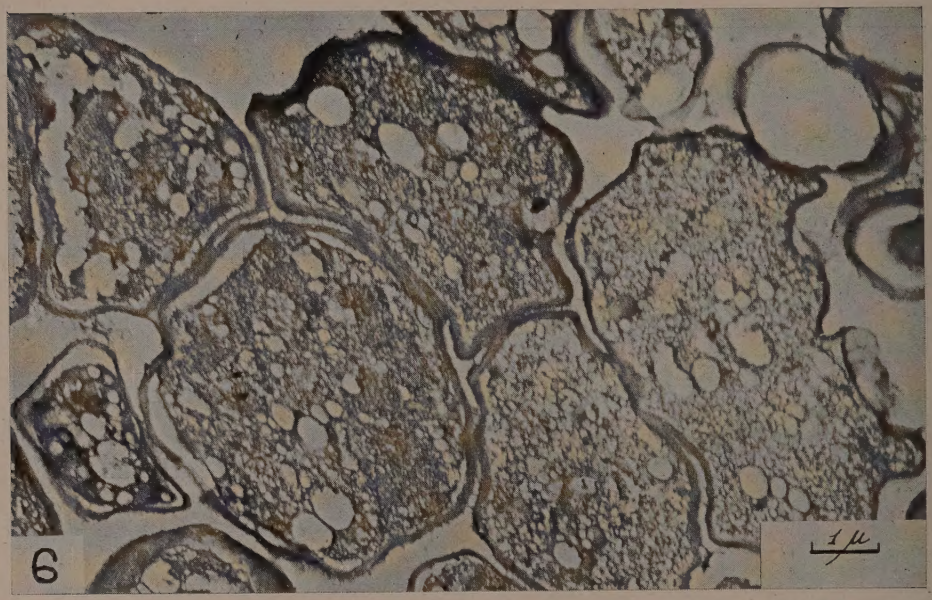
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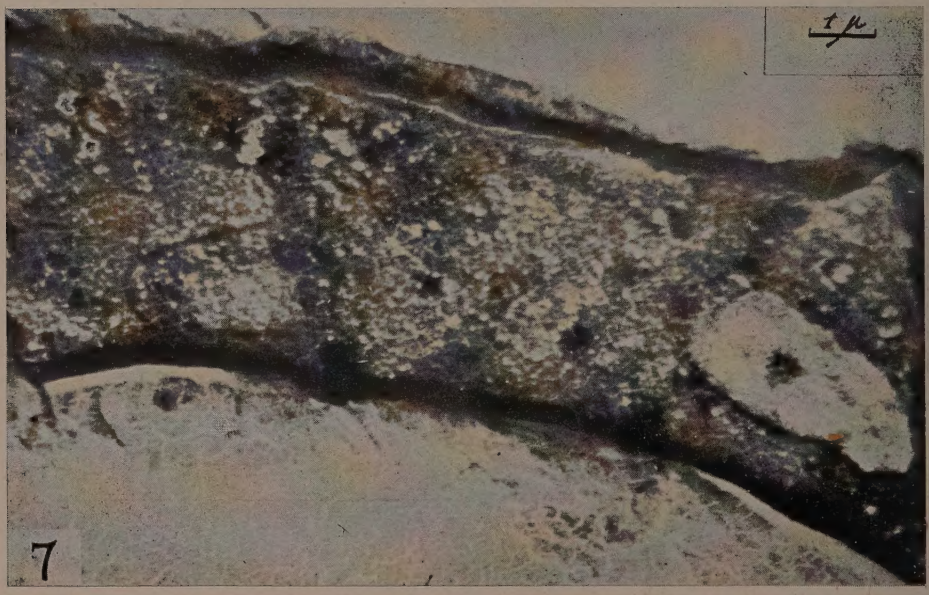
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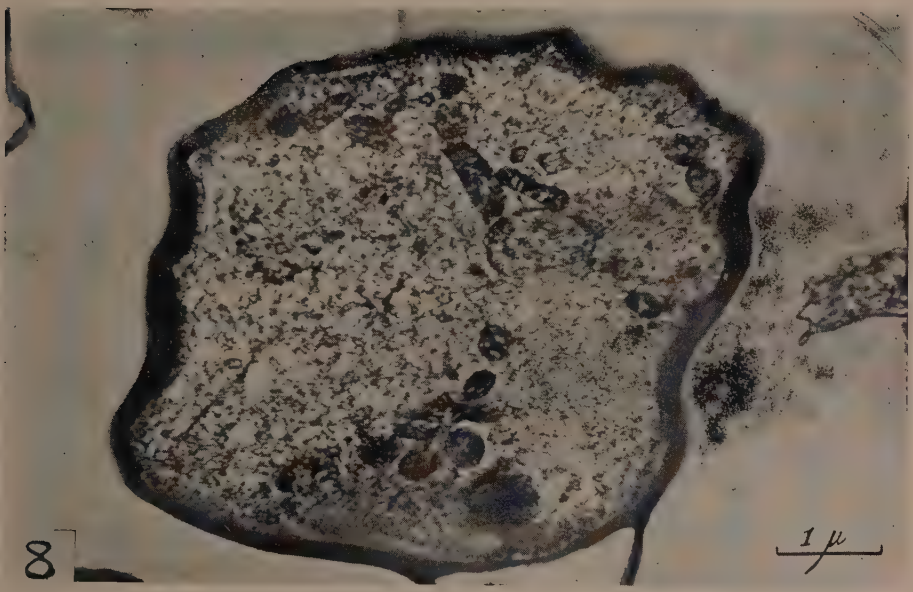
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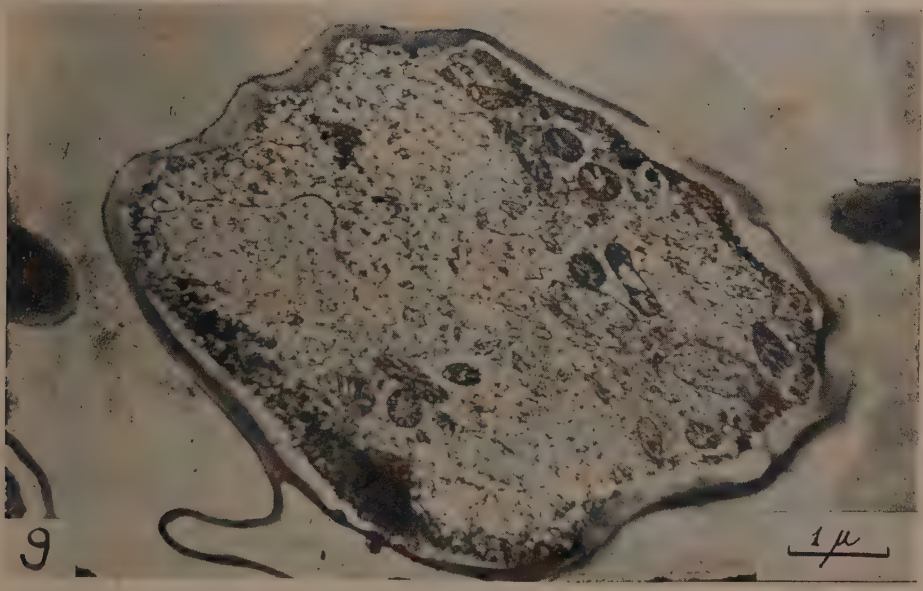
9
1 μ

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STUDIES IN GRAM RUST, *UROMYCES CICERIS-ARIETINI* (GROGN.) JACZ.

H. K. Saksena and R. Prasada

(Accepted for publication, September 16, 1955)

Introductory

The rust of gram caused by *Uromyces ciceris-arietini* (Grogn.) Jacz. is prevalent in Northern India but appears rather late in the growing season in the Indo-Gangetic plain and Bihar. It has also been reported from Bombay and Madras States. The extent of damage caused by the rust has not been actually determined, but judging from the premature death of the infected leaves the loss in out-turn is probably considerable.

So far only the uredo and teleuto stages are known ; the pycnial and aecidial stages have not been observed. Leaves, stems and even the petioles get infected with minute brown pustules which later turn black due to the formation of teleutospores. In nature the rust appears in Delhi in the last week of February when the plants are about four months' old. In places like Karnal which is situated 75 miles north of Delhi and nearer to the foot-hills, the rust breaks out about a month earlier and is known to cause heavy infection.

Apart from a brief account by Butler (1918) and Mehta and Mundkur (1946), there is no information in literature on the life history and epidemiology of this rust. A detailed study of the causal fungus was therefore undertaken and the results are presented here.

Experimental

(i) *Germination of uredospores :* Fresh spores were put for germination in tap water, distilled water and cane sugar solution at 18°C. Since equally good germination was obtained in all cases, only tap water was used in all subsequent tests. At this temperature germination commences in 2½ hours irrespective of light or darkness. There are 4—8 germ pores in each spore but as a rule only one germ tube is produced which grows to 300—400μ in length and is much coiled over itself.

Temperature range suitable for the germination of uredospores was determined. Tests were carried out at different temperatures and records were taken after 24 hours. The results based on an average of three readings are given in Table 1.

TABLE 1

Germination of fresh uredospores at different temperatures

Temperature °C	Percentage Germination	Length of germ tubes (μ)	Width of germ tubes (μ)
5°— 7°	20—25	70—80	7
11°—12°	84—88	190—195.8	7
18°—20°	91.5	260	7
27°	13—15	50	8
30°	6—8.5	28—30	8.5
35°	Nil	—	—
40°	Nil	—	—

These results show that germination of uredospores can take place upto 30°C but the optimum range lies between 11°– 20°C. At 35°C and above the spores do not germinate. This explains why a culture of this rust could not be established at Delhi during the summer months, i.e., April-October, under natural weather conditions.

The uredospores remain viable for 48 weeks when stored at 5°—7°C but are killed within two weeks if kept in the room (average temperature 35°C) on 20th April. All viability is lost if infected gram plants are exposed to 45°C for 72 hours, 40°C for 96 hours and 35°C for eight days This shows that even if a suitable host were present the rust would not oversummer in the plains, because neither the spores would remain viable for long nor would they germinate at temperatures above 35°C. Even between 30° and 35°C the germination, if any, is likely to be negligible.

(ii) *Culture of the rust and incubation period :* A culture of the rust could be easily maintained under natural conditions with the usual technique of inoculation during the winter months upto the middle of April in Delhi. The incubation period at different temperatures was found to be 15 days at 11°—12°C ; 13 days at 17°C ; 11 days at 20°—25°C and 9 days at 25°—30°C. As expected, no infection was obtained at 35°—40°C, since at this temperature the uredospores cannot germinate. Therefore to tide over the unfavourable period, the uredospore dust was put in sealed vials and stored at 5°—7°C where it was found to remain viable for a period of 48 weeks.

Although by this method material could be stored in a viable condition through the summer months, it was obviously not possible to do any laboratory work on this rust for nearly six months, i.e., April to October. In order to overcome this difficulty different methods for culturing the rust were adopted.

As already stated inoculations made during the summer when the temperature in shade was 35°--40°C did not succeed ; also, the

uredospores did not show any germination at this temperature. Obviously, when the spores could not germinate, no infection was possible. Therefore, in the modified method, the seedlings were inoculated and incubated in a moist chamber at 18°C for 48 hours, so that the spores could germinate and infect the leaves. The inoculum showed good germination on the slide which was kept along with the inoculated plants. After 48 hours at 18°C, the plants were transferred to the verandah where the temperature was about 35°C. The rust appeared in 10 days. In this way successive generations of rust cultures could be maintained throughout July, August and September. In May and June, however, when the temperature was above 40°C, this method of rust culture was unsuccessful. These experiments indicate that the temperature which is suitable for the germination of spores bears no relation to the temperature at which pustule formation takes place.

For maintaining rust cultures during May and June it was necessary to keep the host plants at a low temperature (10°- 20°C.) throughout the incubation period. This range of temperature was available only in the cool cabinets under complete darkness but in the absence of light the plants, if grown in water culture, got etiolated in 3-4 days. The possibility of growing gram plants in complete darkness was, therefore, investigated by supplying different concentrations of sugar solutions through the cut end of the stem. The sugar solutions used in these experiments were dextrose, chemically pure sucrose and commercial cane sugar of 1-20 percent concentrations, both sterilized and unsterilized. It was observed that the plant thrived better at lower concentrations, upto 7.5%, than at higher concentrations. Unsterilized 5% cane sugar solution gave the best results and was used in these experiments. Young gram seedlings cut at the soil level and the cut end dipped in sugar solution in small narrow-mouthed bottles, remained healthy for more than three weeks in complete darkness at 18°C. This period was quite sufficient for raising one generation of rust culture. Callus and root formation took place at the cut end in 10-15 days. The leaves of such plants when inoculated produced rust pustules in about 11 days and the inoculum could be multiplied easily in successive generations.

It was found that leaves exposed to diffused light for a short period of 2 hours every morning survived for a week longer than those kept in complete darkness; seedlings detached from the parent shoot towards the evening survived longer in culture than those taken in the morning, and young leaves from 7 to 15 day-old seedlings were found to live longer in culture than the leaves taken from older plants.

Detached leaves have been used successfully for the culture of certain powdery mildews and other rusts (Yarwood, 1946). The method described here has the advantage over other methods in which leaves were floated on the solutions, in that the upper parts of seedlings are not in contact with the solution. Consequently, the rust cultures grow free from saprophytic fungi and the uredospores do not germinate *in situ*.

Development of callus and roots has been observed from the cut end of the stem in 5% and 10% cane sugar solution at 18°C after 10-15 days. With the development of the root system the plant resumed its normal growth. There was a marked elongation of the stem bearing leaf buds at the top. New leaves were produced with the normal green colour in those cases where the plants were exposed to diffused sun light for 2 hours every morning.

Stems cut at different heights above the soil were put in sugar solutions but the root formation occurred only in those plants which were cut upto half an inch above the soil.

By these methods gram plants could be made to live for about three weeks in darkness at 15°—18°C and rust cultures could be maintained throughout the hot summer months when this was not possible in nature. Uredospores produced under these conditions were normal and showed good viability.

(*iii*) *Formation and germination of teleutospores :* In nature, teleutospores are scarce and appear towards the end of April when the crop is ready for harvest. Inspite of various treatments like freezing, thawing, alternate wetting and drying, and use of chemicals, it was not possible to secure the germination of teleutospores collected in April, except in one case where the spores had been soaked in water for 28 days at 20°C and one teleutospore showed germination. It appears that, as in the case of *Puccinia graminis*, teleutospores formed in the plains in April are not viable on account of prevailing high temperature. Unfortunately, teleutospore formation did not take place at lower temperatures (20°—25°C) in gram rust.

Waters (1928) was able to stimulate the formation of teleutospores in different rusts by manipulating the supply of carbohydrates to the detached leaf cultures. In our experiments increase of sugar concentration above 10% in the nutrient solution stimulated the formation of teleutospores in detached leaf cultures.

In some rusts teleutospore-production is more profuse on some host varieties than on others. In *Puccinia triticina* it has been found that teleutospore-formation takes place freely on wheat variety I. P. 114, whereas it is either rare or absent on other varieties (Prasada, 1948). Thirty varieties of gram (N. P. series) were artificially infected. All of them proved to be susceptible and none showed any tendency towards teleutospore production. The formation of teleutospores depends on a variety of factors and further investigation in this direction is necessary.

(*iv*) *Collateral Host : Trigonella polycerata L.*, which is a leguminous weed, has been found to be a collateral host of gram rust and incidentally this appears to be first record of rust on this host. Cross inoculation tests with rusts from the two hosts gave positive results. This finding is important in so far as *Trigonella polycerata* is widely distributed in the hills upto 6,000 feet altitude (Collett, 1921) where conditions for the oversummering of rust are favourable.

Conclusions

From the foregoing it is obvious that the rust is not able to oversummer in the plains in the uredostage. The teleutospores formed on the crop in April have not been shown to be viable, consequently their role in the perpetuation of the rust remains obscure. While more work is required to study the conditions governing the formation and germination of teleutospores, the late appearance of rust in April when environmental conditions are favourable for rust development from October onwards suggests absence of a local source of infection. The wide distribution of the collateral host *Trigonella polycerata* in the hills, where climatic conditions are suitable for the perpetuation of the rust in the uredo-stage during summer, indicates the possibility of rust survival in the hills followed by dissemination to the plains.

The possibility of maintaining rust cultures during the summer months in the plains on plants grown in nutrient solutions has been indicated.

Grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for facilities and encouragement.

Division of Mycology & Plant Pathology,
Indian Agricultural Research Institute,
New Delhi.

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A MOSAIC DISEASE OF RADISH (RAPHANUS SATIVUS L.)

S. P. RAYCHAUDHURI AND P. S. PATHANIAN

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INTRODUCTION

In the vegetable plots at the Indian Agricultural Research Institute a mosaic disease of radish (*Raphanus sativus* L.) was observed in 1951. The characteristic symptoms of the disease are mosaic mottling of young leaves often associated with circular interveinal chlorotic areas (Plate I, Figure 1) which gradually increase in size and finally coalesce to form irregular chlorotic patches. The affected plants are stunted and their leaves reduced in size. Frequently, necrotic lesions appear on the stem, petiole and midrib of the affected plants. In the case when necrotic lesions appear on the mid-rib the leaf first bends outwards and then the mid-rib breaks at the necrotic region so that a part of the leaf collapses. (Plate I Figure 2). The disease incidence varied from 6 to 8 per cent in the field.

The disease is readily transmissible to radish by sap-inoculation, but not through the seed. The host range and properties of the virus have been studied and the account is briefly presented here.

EXPERIMENTAL

Experimental plants were raised in the greenhouse from seed obtained from the Division of Botany as well as from commercial seed stores. All inoculations were done by the leaf rubbing method using carborundum as an abrasive which facilitated transmission of the virus. Standard extract (1 c.c. of distilled water added to each gram of diseased leaf material) was used in all the inoculation experiments except where otherwise mentioned.

(a) *Host range of the virus*:—Inoculations were made on 36 species of plants belonging to *Cruciferae, Solanaceae, Cucurbitaceae, Leguminosae, Gramineae, Malvaceae, Chenopodiaceae* and *Compositae*. The virus was found to have a restricted host-range and infected only the plants belonging to *Cruciferae*, viz. *Raphanus sativus* L., *Brassica rapa* L., *B. campestris* L. var. Sarson Prain T71 and T87, *B. campestris* var. toria Duthid and Fuller T. 18, *B. nigra* (L.) Koch. T253, *B. alba* Rabenh. T. 93 and *B. juncea* (L.) Coss. T 187. *Brassica oleracea* L., *B. oleracea* var. *botrytis* L. and *B. oleracea* var. *capitata* L. did not show any visible symptoms of the disease, but the virus could be recovered from the inoculated areas only proving thereby that the virus does not multiply in these hosts,

(b) *Symptomatology*:—*Raphanus sativus*—visible symptoms appeared 8-10 days after inoculation in the form of vein-clearing followed by chlorotic spots on the leaves. After 15-20 days, the

PLATE I

Fig. 1—Symptoms of the mosaic disease on radish under field conditions.

Fig. 2—Necrotic symptoms on radish.

Fig. 3—Symptoms on radish under green-house conditions.

Fig. 4—Symptoms on *Brassica rapa*.

PLATE II

Fig. 5—Symptoms on *Brassica campestris* var. *Sarson* T87.

Fig. 7—Fruits of *B. campestris* var. *Sarson* T87-A. healthy ; B. diseased fruits.

Fig. 6—Symptoms on flowers and fruits of *B. campestris* var. *Sarson* T87.

chlorotic spots spread all over the leaf surface. Later, these spots increased in size and coalesced finally forming large irregular chlorotic patches with mosaic (Plate I Fig 3). Frequently, necrotic lesions appeared on the midrib of the leaves and stem. The necrotic lesions at the base of the stem gradually spread in a linear fashion towards the apex and affected the leaves ultimately killing them.

Brassica rapa—The first visible symptoms appeared in the form of circular chlorotic lesions on the inoculated leaves 8-10 days after inoculation (Plate I, Figure 4). Systematic infection appeared when light and dark green patches were formed on the young leaves. Necrotic lesions were also formed on the stem as well as the midrib of the leaves as in radish.

Brassica campestris var. *Sarson T71* and *T87*; *B. compestris* var. *toria T18*—Symptoms on these hosts were practically the same excepting that they were more severe on *B. campestris* var. *Sarson* T87. Circular chlorotic areas developed on inoculated leaves 7-10 days after inoculation. The infection became systemic after 10-15 days when faint mottling associated with severe necrosis on the stem was observed on diseased plants. The necrotic areas spread from the stem to the leaves and midribs finally killing the leaves (Plate II, Figure 5). Affected leaves were much reduced in size. The flowers borne on infected plants were fewer in number and all of them did not open normally (Plate II, Figure 6). A few fruits that developed were malformed, variously twisted, much reduced in size and exhibited necrotic spots all over the surface (Plate II, Figure 7).

Brassica nigra (*T 253*):—The symptoms appeared as necrotic lesions in 8 to 10 days on the inoculated leaves. After 10-15 days the disease became systemic, and necrosis spread over the leaves and stem while chlorotic lesions were formed on the youngest leaves. The leaves were extremely reduced and much twisted so as to give the plant a stunted and bushy appearance. Occasionally the necrotic areas spread from the base of the stem towards the apex thereby killing the entire plant.

Brassica alba (*T 93*):—Necrotic lesions developed on the inoculated leaves, 7-10 days after inoculation.

(c) *Properties of the virus*:—Thermal death point of the virus was determined by heating the standard extract in water bath at various temperatures for 10 minutes. The virus withstood heating at 85°C but was rendered innocuous when heated to 90°C for the same period.

The virus in crude juice withstood a dilution of 1 : 10,000,000. The virus was found to retain infectivity for 17 days at 17-22°C but was rendered innocuous after 19 days at the same temperature. At a temperature range of 6-8°C however, the virus retained infectivity after 101 days of storage.

Discussion

The virus under consideration is of great economic importance as it affects many oilseed crops producing in them severe necrosis

resulting in death of the plants. It differs from turnip mosaic (*Marmor brassicae* H.), radish mosaic (*Marmor raphani* H.) and the one belonging to turnip virus 1 group (Chamberlain, 1936; Tompkins, 1939; Berkeley and Weintraub, 1952) in host range and properties. Although cauliflower mosaic (*Marmor cruciferarum* H.) (Tompkins, 1937) resembles in host range the virus under consideration, it differs in properties. Larson and Walker (1939, 1941) investigated the mosaic and ring necrosis of cabbage while Walker, Le Beau and Pound (1945) classified cabbage virus A, cabbage black ring and and ring necrosis viruses in turnip virus 1 group, and the cabbage virus B and broccoli virus in cauliflower virus 1 group respectively. These viruses differ in host range and properties from those of radish mosaic virus described here. Severin and Tompkins (1948) reported a cauliflower mosaic virus which produces prominent symptoms of vein-clearing, vein-banding, mottling, distortion and necrotic lesions on cauliflower, differing thereby from the virus reported herein. Also, radish mosaic of Delhi differs in host range from those of the strains of cucumber mosaic occuring on crucifers (Pound and Walker, 1948). Sylvester (1953) recently reported a virus occuring on *Brassica nigra* belonging to radish virus 1 group, which produces local lesions on *B. juncea* but differs in properties of this virus.

The investigations were initiated by Dr. R.S. Vasudeva, Head of the Division of Mycology and Plant Pathology, I.A.R.I. The authors are indebted to him for his constant interest and helpful criticism.

Summary

A virus causing mosaic disease of radish, host range of which is restricted to Cruciferae, has been described. It causes necrosis in some of the common oil seed crops.

The virus withstands exposure to 85°C for 10 minutes but not 90°C., dilution to 1 : 10,000,000, and 17 days and 101 days storage at 17°-22°C. and 6°-8°C., respectively. The disease is not transmissible through the seed.

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A NEW LEAF SPOT DISEASE OF *PIPER BETLE* IN WEST BENGAL

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INTRODUCTION

Piper betle L. grown extensively in West Bengal suffers from a number of diseases. McRae (1934) mentioned three important diseases of *Piper betle* L., namely underground stem rot caused by *Sclerotium rolfsii* Sacc., foot rot caused by *Rhizoctonia solani* Kuhn and wilt caused by *Phytophthora parasitica* Dast. Roy (1948) described an anthracnose disease caused by *Colletotrichum dasturii* Roy. Hector (1921) mentioned that a species of *Fusarium* was also isolated along with other fungi from betel vines, showing yellow discolouration and wilting of leaves and blackish brown decay of the roots. Padwick (1942) isolated a species of *Fusarium* from the stipules, stems and leaves of "foot rotted" plants.

During the course of a recent investigation conducted to find out the prevalence of various diseases of *pan*, a species of *Fusarium* was found associated in many cases with fungi attacking leaves, particularly with *Colletotrichum dasturii*. In a number of cases the *Fusarium* sp. only could be isolated from spotted leaves.

Leaves from where *Fusarium* alone could be isolated are characterised by the presence in the central region of a large circular spot (Fig. 1) with a number of broad and distinct concentric zones of alternate chocolate brown and light brown colour. The spots are large, well developed and may often attain a diameter of 4—6 cm. while the individual zone may be 0·5—1 cm. in width. There may be two to three spots in a leaf. The attacked stipules show blackening often followed by rotting. Zonate spots are not usually found.

Study of the organism.

The organism was isolated in the usual way from the affected portions of leaves and stipules. The fungus *Fusarium* isolated was purified by single spore technique. Large scale isolations of the organism showed that the *Fusarium* is often associated with other fungi particularly *Colletotrichum dasturii*.

The fungus, after its purification in culture was grown in different media and plant tissues to study the growth, pigmentation, production of chlamydospores and conidia and the variation in the morphological characters of the conidia. Cultures under study were kept at temperatures which varied from 26—28°C. For noting of

Fig. 1—An attacked leaf of *Piper betle* showing characteristic symptoms.

colour, Ridgway's "Colour standards and colour nomenclature" was consulted. The details of macroscopic growth characters are presented in Table 1.

TABLE 1

Macroscopic growth characters of the fungus in different media.

Medium	Colour of the hyphae	Colour of the growth of the substrate	Colour of the medium
Potato dextrose agar (with 2% dextrose)	aerial hyphae scanty, pale pinkish in colour.	pale cinnamon pink, powdery spore mass present on the surface.	pale pinkish cinnamon,
Potato dextrose agar (with 5% dextrose)	aerial hyphae rare with pinkish hue.	pale cinnamon pink, powdery spore mass present.	light pinkish cinnamon.
Maize agar	aerial hyphae moderate, white touched with pinkish hue.	pale cinnamon pink, powdery spore mass present.	pale cinnamon pink.
Brown's synthetic agar	aerial hyphae moderate, white with pinkish hue.	white with pinkish hue mixed with yellowish specks.	pale pinkish buff.
Steamed rice	aerial hyphae moderate, orange pink.	gerardine pink.	baryta yellow.
Potato plug	aerial hyphae moderate, white touched with pinkish hue.		

Details of conidia and chlamydospores formed by the fungus on different media are given below.

POTATO DEXTROSE AGAR (containing 2% dextrose)

Conidia scattered throughout the aerial surface of the mycelium as powdery masses. Sporodochia absent. Conidia pinkish in colour, both micro- and macro-conidia typically spindle shaped with tapering ends or in macro-conidia, one end may be slightly curved, apedicellate,

micro-conidia 0-2 septate, macro-conidia 3-5 septate, occasionally 6 septate measuring :

0-septate— 1%	6.4×1.6μ	
1-septate— 5%	12.8×1.7μ	(11.2—14.4×1.6—2.4μ)
2-septate— 5%	15.3×2.5μ	(12.8—20.8×1.6—3.2μ)
3-septate—28%	22.5×3μ	(16 - 28.8×2.4—4μ)
4-septate—37%	26.5×3.4μ	(19.2—30.4×2.4—4.8μ)
5-septate—22%	31×3.2μ	(25.6—35×2.4—4μ)
6-septate— 2%	35.2×4μ	

Chlamydospores practically absent.

Potato Dextrose Agar (containing 5% dextrose)

Conidia scattered over the aerial hyphae as powdery masses. Sporodochia absent. Conidia in mass light pinkish in colour, apedicellate, both micro- and macro-conidia typically spindle shaped, broadest at the centre with both ends tapering and pointed ; macro-conidia occasionally curved at the ends, micro-conidia 1-2 septate, macro- conidia 3 septate measuring :

1-septate 33%	12×2μ	(8—16×1.6—3.2μ)
2-septate 15%	15.5×2.6μ	(12.8—19.2×2.4—3.2μ)
3-septate 52%	20.2×2.8μ	(14.4—25.6×2.4—3.2μ)

Chlamydospores intercalary, oval in chains, hyaline, smooth in the beginning, may become finely spiny at maturity (measuring 8-11.2 ×4.8-6.4μ).

Maize Agar

Conidia scattered in loosely arranged hyphae, pinkish in mass, micro- and macro-conidia both typically spindle shaped with both ends pointed, apedicellate or may be pedicellate micro-conidia 1-2 septate, macro-conidia 3 septate measuring :

1-septate 18%	13.2—2.2μ	(9.6×14.4—1.6×2.4μ)
2-septate 18%	16×2.6μ	(12.8—17.6×2.4—3.2μ)
3-septate 64%	20×3μ	(16 - 25.6×2.4—3.2μ)

Chlamydospores intercalary, oval, in long chains, hyaline, smooth but later on finely spiny, measuring 6.4-9.6×4.8-8μ.

Brown's Synthetic Agar

Conidia scattered in aerial mycelium as powdery masses. Sporodochia absent. Conidia pinkish in mass both micro- and macro-conidia typically spindle shaped, broadest at the centre with tapering ends, macro-conidia may be curved at both ends, but abruptly

pointed, micro-conidia 1-2 septate, macro-conidia 3-6 septate, measuring :

1-septate	13%	15.7—2.4μ	(12.8—20.8×1.6—2.8μ)
2-septate	10%	18×3μ	(16—20.4×2.4—3.2μ)
3-septate	51%	24×3.4μ	(17.6—30.4×3.2—4.8μ)
4-septate	8%	29.2×4.4μ	(24—33.6×3.2—4.8μ)
5-septate	8%	36.3×4.5μ	(30.4—40×4—4.8μ)
6-septate	10%	42.2×4.5μ	(35.2—51×3.2—4.8μ)

Chlamydospores intercalary, round, oval or irregular in shape, in long continuous chains, hyaline, smooth, may become spiny at maturity, measuring 6.4-9.6×6.4-8.4μ.

Steamed Rice

Conidia loosely arranged in hyphal mat. Sporodochia absent. Conidia in mass pale pinkish both micro- and macro-conidia typically spindle shaped, in macro-conidia one or both ends may be slightly curved, micro-conidia 1-2 septate, macro-conidia 3-5 septate, one or more intermediate cells of macro-conidia may be transformed into chlamydospores, measuring :

1-septate	14%	14×2.7μ	(11.2—1.6×2.4—3.2μ)
2-septate	15%	18.3×3.3μ	(14.4—20.8×3.2—4.8μ)
3-septate	59%	24.5×3.5μ	(19.2—30.4×2.4—4μ)
4-septate	7%	28.4×3.6μ	(20.8—30.4×3.2—4μ)
5-septate	5%	35.3—3.7μ	(29.2—48×3.2—4.8μ)

Chlamydospores intercalary, in mycelium and conidia round or oval or irregular in size, in long continuous chains, hyaline, smooth and measuring 8.4-9.6×6.4-9.6μ.

Potato Plug

Conidia scattered loosely in hyphal mat, pinkish in mass, may be hyaline, both micro- and macro-conidia spindle shaped, with both ends tapering and pointed ; micro-conidia 1-2 septate, macro conidia 3-6 septate, measuring :

1-septate	5%	14.4×2.4μ	(16.2—17.6×2.4μ)
2-septate	5%	16.8×3.2μ	(16—17.6×3.2μ)
3-septate	52%	23.4×3.4μ	(19.2—27.2×2.4—4μ)
4-septate	27%	30×3.5μ	(28.8—32×3.2—4μ)
5-septate	9%	32.4×3.4μ	(25.6—36.8×3.2—4μ)
6-septate	2%	32×4.8μ	

Chlamydospores intercalary, round, oval or irregular in shape, in long chains, hyaline, smooth, may be spiny at maturity, measuring, 8—11.2×7.6—8.4μ.

Presence of whitish to pink coloured aerial mycelium, spindle shaped micro-conidia 0-2 septate, intercalary chlamydospores in mycelium and in conidia, and absence of sporodochia or pionnotes, place the fungus under section Arthrosporiella according to the system presented by Woolenweber and Reinking (1935).

The fungus is identified as *Fusarium semitectum* Berk. and Rav., because of straight, spindle shaped macro-conidia and its measurements.

Pathogenicity Tests

To test the pathogenicity of the fungus, fresh betel-vines were collected. They were made into small bits, each consisting of an apical bud, two leaves and a comparatively long stem with some adventitious roots. The cut end was placed in nutrient (Knop solution) solution in 500 c. c. conical flasks. The whole twig was kept in position with the help of loose cotton plug. The twigs were then sprayed with aqueous suspension of spore from fifteen days old culture and kept inside bell-jars to keep them under proper humidity. A set of fifteen flasks was used for the purpose of inoculation, while a set of same number was kept as control. Within 48 hours, blackening and rotting of stipules were observed. On the leaves, small spots with faint zonations were also noticed. The twigs dried up after a period of 5-6 days in all the cases, but in the control, no rotting of stipules or production of spots was noted. Reisolations made in all the cases of attack (rotting of stipule and formation of leaf spot) revealed the presence of the same fungus *Fusarium semitectum.*

Fusarium semitectum has been reported from India previously by Mitra (1934), who isolated the organism from greyish spots on a green fruit of *Citrus medica.*

Summary

1. *Piper betle* suffers from a leaf spot caused by *Fusarium semitectum*. The organism alone may produce the characteristic circular patches on the leaves. It may also be associated with other fungi particularly *Colletotrichum dasturii.*

2. *Fusarium semitectum* was grown on a number of media and its cultural characters studied.

3. Pathogenecity of the organism has been established.

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FUSARIUM BLIGHT OF GUAR

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Guar (*Cyamopsis tetragonoloba* Taub.) is an important leguminous crop grown extensively in Gujarat. It is primarily used as fodder for cattle, but pods of certain varieties are also used as vegetable. Among the varieties grown normally Patel, Patel and Patel (1952) have identified two distinct types, *viz.*, hairy and glabrous. Hairy type is more suitable as cattle feed while the glabrous type is more suitable for use as vegetable since its pods are very tender. It suffers from numerous diseases, amongst which root rot caused by *Fusarium caeruleum* (Lib.) Sacc. has been reported to be serious by Prasad (1944) in Sind and by Singh (1951) in U.P. Powdery mildew has been reported from every tract wherever it is grown. Uppal, Patel and Kamat (1934) have recorded *Phoma sp.* on it, but it seems that determination of species has not been done so far.

In September 1949, a peculiar disease of guar was found on the Farm attached to the Agricultural Institute, Anand. The disease was in the nature of a blight affecting the leaves and stems of the plant and was characterised by the appearance of black streaks near the axils of the leaves, (Fig. 1). In nature the symptoms take the form of a blight of aerial parts of *guar*. The disease started as a black streak on stem which extended both upwards and downwards as recorded by Prasad and Desai (1952). The pedicels of the leaves were also affected and gradually the shoots. In very severe cases of attack, the entire plant presented a wilted appearance. These symptoms appeared only on hairy varieties of *guar*. Examination of transverse and longitudinal sections of affected tissue revealed the presence of intracellular fungus hyphae within the parenchymatous tissues. In advanced cases, some hyphae were found to be present in the vessels as well.

Extent of Damage

As the disease was observed to be of a very serious nature in 1949 and appeared again in September 1950, a survey of the *guar* crop in the different sections of the Institute was undertaken. Random samples were taken and the number of diseased plants counted. As the Institute has an 800 acre Farm divided in various sections it provided an opportunity of assessing damage on a large area. The data collected are recorded in Table 1 giving an idea of the damage caused by the disease.

Fig. 1

Healthy Diseased

Symptoms in nature of the blight of guar (Hairy Type)

TABLE 1

Percentage infection of guar blight in different sections of the Institute

Section.	Plot	Total No. of plants examined	No. of plants infected	Average infection percent.
College.	A	510	290	
,,	B	496	280	58.6
,,	C	504	310	
Horticulture	A	490	70	
,,	B	512	56	10.0
,,	C	495	32	
Commercial Farm	A	492	204	
,,	B	504	300	50.0
,,	C	509	248	

RELATION OF ENVIRONMENT TO DISEASE

The disease was first observed during the later part of the monsoon in 1949. The incidence of the disease was reduced as the monsoon withdrew, but in the year 1950, it again appeared in a virulent form. This suggested that there was some relationship between the climatic conditions and the appearance of the disease. To understand the nature of this relationship, a study of rainfall, temperature and humidity was undertaken for a period of five seasons, *viz.* 1949-1953.

1. *Rainfall.*

In 1949 and 1950, as shown in Table 2, it rained regularly during the month of September, while during 1951 and 1952, there was no rain in the corresponding period. In September 1953, there was good rain in the first fortnight, while no rain fell in the second. The average rainfall per week during the last five years is recorded in Table 2. It can be seen that rainfall figures during the year 1953 are comparable to that for 1949 and 1950 except that there was no rain during the third and fourth weeks of September. The absence of rain during this period reduced the atmospheric humidity during the early part of October and this fact was possibly responsible for checking the spread of the disease after an initial start.

TABLE 2

Average daily rainfall by weeks in the year 1949 to 1953

Month	Week.	1949	1950	1951	1952	1953
July	1st.	0.26	1.24	0.30	0.40	0.30
	2nd.	0.31	1.55	0.00	0.00	0.37
	3rd.	0.90	0.93	0.12	0.25	0.03
	4th.	0.34	0.72	0.93	2.30	0.13
August	1st.	0.34	0.15	0.13	0.48	0.47
	2nd.	0.24	0.03	0.37	0.09	1.40
	3rd.	0.05	0.00	0.36	0.00	1.06
	4th.	0.05	0.00	0.00	0.00	0.41
September	1st.	0.03	0.20	0.00	0.00	0.41
	2nd.	0.60	0.17	0.00	0.00	0.13
	3rd.	0.18	0.13	0.00	0.00	0.00
	4th.	0.18	0.00	0.00	0.00	0.00

TABLE 3

Maximum daily temperature by weeks in the years 1949 to 1953.

Month.	Week.	1949	1950	1951	1952	1953
July	1st.	101.5	100.0	100.5	100.0	97.0
	2nd.	99.0	85.0	100.5	100 0	92.0
	3rd.	93.5	95.0	99.0	98.0	92.0
	4th.	94.0	88.5	101.0	90.0	98.0
August	1st.	90.5	90.5	96.0	83.5	94.0
	2nd.	92.5	90.0	90.0	91.0	91.0
	3rd.	93.5	93.5	91.5	92.5	89.0
	4th.	97.5	94.5	94.5	94.5	93.0
Septembet	1st.	98.0	93.0	97.0	95.0	95.0
	2nd.	95.0	95.0	97.0	94.0	95.0
	3rd.	95.0	92.0	98.0	95.0	95.0
	4th.	97.0	95.0	103.5	98.0	96.0

2. *Temperature.*

The temperature during the months of September in the years 1949, 1950 and 1952 was almost similar with a normal maximum of 95°F. From the record of incidence of the disease, it can be readily seen that the disease was widespread during the years 1949 and 1950, and was completely absent in the year 1951. Although the temperature was much lower in 1952 and 1953, the disease was not so severe.

It was completely absent in 1952 while in 1953, it appeared in the beginning of September and it was feared that the year would record the same percentage of attack as those of 1949 and 1950, but rains failed during the later part of September and consequently the disease did not make any progress. Maximum temperatures in a week are recorded in Table 3.

3. Humidity

The disease appeared to be worse in the later part of September in the years 1949 and 1950. Preceding this period, the rainfall was frequent and the humidity had never gone below 90% while in the years 1951 and 1952, during the same period the humidity was below 80%. By comparing the figures of incidence, it appears likely that the progress of the disease was helped by high humidity

Even during the years 1949 and 1950, the progress of the disease was checked when the percentage of humidity went below 90%. During the year 1953, the disease made its appearence in the beginning of September when humidity was high. As no rains occurred after the second week of September, 1953, the progress of the disease was checked, The average daily humidity by weeks has been recorded in Table 4.

TABLE 4

Average daily humidity by weeks in the years 1949 to 1953.

Month.	Week.	1949	1950	1951	1952	1953
July	1st.	82.7	81.0	79.0	78.0	83.0
	2nd.	92.0	94.5	79.0	78.0	83.0
	3rd.	89.0	93.0	91.0	84.5	89.5
	4th.	93.0	92 0	95.0	96.0	83.0
August	1st.	92.5	90.5	95.0	92.5	92.5
	2nd.	91.0	87.0	86.0	86.5	91.5
	3rd.	91.0	84.5	91.0	82.5	93.0
	4th.	91.0	85.0	79.0	83 0	89.0
September	1st.	91.0	85.0	79.0	83.0	89.0
	2nd.	95.0	93.5	79.0	81.0	92.0
	3nd.	95.0	94.0	79.0	75.0	92.0
	4th.	95.0	92.0	72.0	67.0	83.0

4. Age of the plant.

Guar, a semi-*rabi* crop, generally sown in August and September, is used principally as a forage crop, while the crop sown in earlier months is used for vegetable purposes. Observations were taken on the incidence of the disease in plots sown on different dates. It was observed that early-sown crop was free from the disease, while the

later sown was heavily infected. Generally, the highest percentage of infection was observed in October, the earlier sown crop escapes infection as it matures earlier. The later sown crop is in a proper condition for the attack of the disease (Table 5).

TABLE 5

Effect of sowing date on severity of infection

Date of sowing.	Date on which observation was taken.	No. of plants examined.	No. of infected plants.	Infection percent.
20-8-1949.	28- 9-1949	512	56	11
4-9-1949.	29- 9-1949	492	204	40
5-8-1950.	30- 9-1950	504	—	—
17-8-1950.	30- 9-1950	495	32	6
5-9-1950.	1-10-1950	504	310	62

ISOLATION

Samples of stem, petiole and pods were collected from diseased plants. Small pieces were cut and were surface sterilised and transferred aseptically to agar slants. Isolations were made at different intervals during the years the disease was observed. A very large number of isolations were made numbering approximately 500 and different types of fungi were obtained and they were roughly classified into sixteen groups. The groupings were made on the basis of presence and absence of aerial mycelium, sporodochia, sclerotia, pycnidia and discolouration of the media. They are listed in Table 6.

TABLE 6

Different groups of cultures obtained from isolations.

Group No.	No. of cultures.	Mycelium Characters.	Fruiting bodies.	Remarks.
1	6	Light pink. Profuse on P. D. A. woolly growth.	Sporodochia not seen.	Discolouration of the medium. Pinkish to bluish.
2	7	White, profuse mycelium on P. D. A. some aerial growth.	do	No discolouration of medium.
3	13	White, profuse aerial mycelium.	do	do

TABLE 6—(*contd.*)

Different groups of culture obtained from isolations.

Group No.	No. of cultures.	Mycelium Characters.	Fruiting bodies.	Remarks.
4	3	Dull white mycelium.	Sprodochia not seen.	Discolouration of the medium. Pinkish to bluish.
5	3	Light pink with slight bluish mycelium. Woolly growth.	do	Slight discolouration of the medium.
6	4	Pink mass of mycelium.	do	No discoloration of medium.
7	8	White profuse aerial mycelium. Aerial mycelium in mass.	Sporodochia not seen.	No discolouration of medium.
8	5	Dull white scanty mycelium.	Dull white sporodochia.	Discolouration of the medium Dark brown.
9	8	Dark olive green to black mycelium.	Pycnidia round and large.	Medium turned black.
10	1	Dull white scanty mycelium.	Dull white sporodochia. More at the centre.	Medium turned dark brown.
11	10	White to olive green mycelium.	Sclerotial bodies.	Medium turned black.
12	8	Mycelium dull white woolly growth.	Scattered perithecia.	No change in colour of medium.
13	3	Light pink with bluish in the centre on P. D. A. woolly growth.	—	Discolouration of medium.
14	4	Mycelium dull, white woolly growth.	Perithecia in rings.	No change in colour of medium.
15	3	Dull white to cream coloured mycelium, dense mass.	—	do
16	12	Scanty pink mycelium submerged in the medium.	Sporodochia pink through out.	Discolouration of medium pink.

Pathogenicity Trials.

Sterilized earthen pots (9″×8″) were filled with soil sterilized at 30 lb. pressure for an hour. *Guar* seeds were sterilized in 0.1% mercuric chloride solution before sowing. Seeds were sown on the 3rd April 1951. Spore suspensions from each of the sixteen groups of isolates were prepared separately and atomised on 26th April 1951, when the seedlings were about 3 weeks old. The seedlings were inoculated in the evening. Control pots were sprayed with sterilized water. After atomizing spore suspensions, the plants were sprayed with water three times a day. In spite of these precautions, the disease failed to appear as the weather during the period was very dry. The experiment was repeated on the 16th August 1951, when the weather was ideal. The humidity was around 90% and the temperatures were not as high as found in April. The symptoms of the disease could be seen on the leaves one week after inoculation.

Two methods of infection were followed, viz., the seed was dipped in spore suspension of different isolates and in second, the spore suspension of each isolate was atomized on three weeks old seedlings. Out of the total of 16 isolates only three were found to be parasitic and these were 1, 3 and 16.

The infection in the early stages was characterised by the development of a few black streaks on stems, but the progress of the disease was checked due to complete failure of rains and sudden increase in temperature as can be seen in Fig. 3. On reisolation from the affected plants, the isolates 1, 3 and 16 were obtained in the culture.

These trials were repeated again during the rainy season of 1952 with similar results. From previous experience, this period was found to be the best as humidity was high and temperature was comparatively low. These experiments conclusively proved that only three out of sixteen isolates were pathogenic on *guar*.

Host Range

In order to determine whether the parasite was able to infect any host other than *guar* generally found during the same season, the following plants, raised in pots were selected for cross infection studies.

Cyamopsis tetragonoloba Taub. (Two types viz. hairy and glabrous.); *Crotalaria juncea* L., *Crotalaria retusa* L., *Gossypium herbaceum* L., *Cajanus cajan* (L), Millsp.

Isolates No. 1, 3 and 16 found to be pathogenic in the earlier experiments were used for inoculation. The seeds of the above eight hosts were dipped in spore suspension of the above three isolates and were sown in pots filled with sterilized soil. The experiment was started on the 16th August 1952. After the plants were three weeks old, spore suspension of the three isolates were atomized on each. Symptoms of the disease appeared on the hairy variety of *guar* nearly

ten days after inoculation. As the conditions were favourable, the disease made good progress and characteristic black streaks were observed. All the three isolates failed to infect any host other than the hairy type of *guar*. The isolates 1, 3 and 16 were reisolated from the infected plants. The experiment was repeated in 1953 with similar results.

Identification of Causal Organisms

In order to identify the causal organism, the morphological characters of the three isolates (1, 3 and 16) were studied on potato dextrose agar and oat meal agar. The presence of micro and macroconidia and typical chlamydospores placed the three isolates in the genus *Fusarium*. The characters of the three isolates are recorded below :—

Culture 1.

Mycelium profuse, pale pink in mass, hyphae hyaline septate and 2.4μ to 2.6μ in thickness, microconidia formed freely in aerial mycelium, relatively abundant, ovoid, oblong or irregularly ellipsoid, mostly 0-septate, macroconidia formed below the mycelial growth, hyaline, thin, long, sickle shaped with 2 to 5 septa, oil globules present, peculiar notch at the end, chlamydospores both intercalary and terminal present, sporodochia and pionnotes absent.

Conidia, 0-4 septate. (Fig. 2).

0-Septate	4.8—32.0×2.4—4.0μ
1-Septate	19.2—22.4×4.0—4.8μ
2-Septate	22.4—25.2×4.0—4.8μ
3-Septate	22.4—32.0×4.0—4.8μ
4-Septate	32.0×4.8μ

Culture 3.

Growth good on potato dextrose agar, woolly, felt like white to dull white, mycelium hyaline, septate and 1.6μ to 2.4μ in thickness, microconidia moderately formed, free aerial mycelium, ovoid, oblong or irregularly ellipsoid, mostly 0-septate, macroconidia absent, chlamydospores both intercalary and terminal present, sporodochia and pionnotes absent.

Conidia, 0-1 septate, (Fig. 2)

0-Septate	6.4—22.4×3.2—4.8μ
1-Septate	8.0—25.6×4.0—6.4μ

Culture 16.

Submerged mycelium, pale pink in mass, scanty, hyphae hyaline, septate and 2.4μ to 3.2μ in thickness, microconidia absent, macroconidia present, hyaline, 0 to 5 septate, long sickle shaped, peculiar

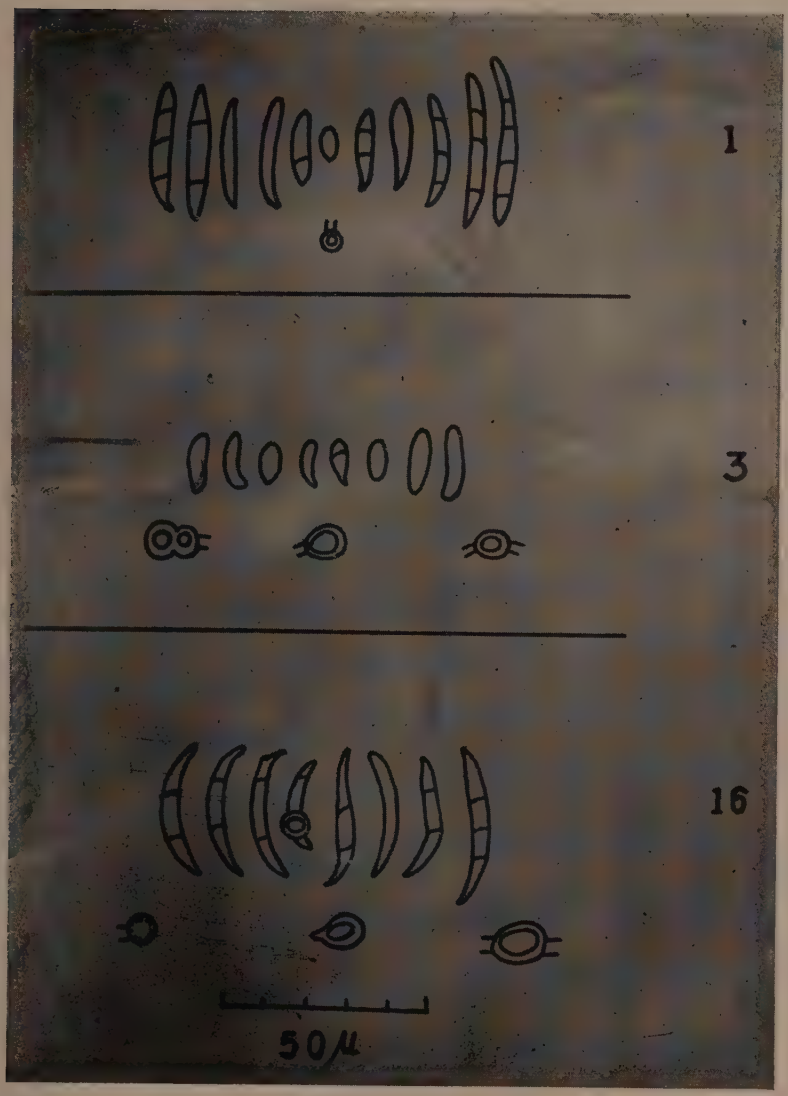

Fig. 2

Conidia of cultures No. 1, 3 and 16.

notch at the end. chlamydospores intercalary and terminal present, pionnotes absent, sporodochia present, pale pink in colour.

Conidia 0-6 septate (Fig. 2).

0-Septate	27.3—46.8×3.9—5.8μ
2-Septate	27.3—39.0×2.9—4.9μ
3-Septate	31.2—58.5×3.9—4.9μ
4-Septate	23.4—46.8×3.9—4.9μ
5-Seprate	51.7×3.9μ
6-Septate	58.8×3.9μ

The above three cultures were characterised by the presence of white or buff to orange coloured mycelium, spindle or oval shaped microconidia, delicate, sickle shaped macroconidia with occassional hooks at the bases and tapering at the other end. These characters place the three strains in section *Liseola* of the genus *Fusarium* according to Wollenweber and Reinking (1935).

Microconidia either combined or in false clusters, yellowish to rose-like, aerial mycelium dispers, one or two-celled, spindle or egg shaped. They are constricted at the basal cell and narrow at the other end, sometimes are somewhat hooked, bent at the base. Macroconidia are distinct or feeble. They are either scattered or in sporodochia. Their colour in mass is bright cream to bright orange. These detailed characters agree with those of *Fusarium moniliforme* Sheld. according to Wollenweber and Reinking (1935).

Snyder and Hansen (1945) have reduced the section to a single species *Fusarium moniliforme* (Sheld) emend. Snyd. et. Hans. We agree with Snyder and Hansen. There are sufficiently wide differences in morphology of the three isolates and on this basis, they could be classified into three species. It is difficult to imagine that three species would cause the same disease on one and the same host. It would be preferable to treat them as clones within one species.

The three isolates were tried for cross inoculation on other hosts. In repeated trials, they failed to cause any symptoms on other hosts. It shows that these isolates are specialised in their parasitism on *guar* only. Snyder and Hansen (1940, 1941 and 1945) have stated that generally specialisation of parasitism is not found in *F. moniliforme* as in *F. oxysporum* and *F. solani* and thereby they have not proposed any trinomial for this species. We are at present not proposing any trinomial for this species as it would require some more work with other isolates of *F. moniliforme.*

Summary

A new and very destructive disease of *guar* appeared during the month of September 1949 and 1950. It was characterised by the appearance of black streaks near the axils of the leaves and the streaks extended both upwards and downwards.

A survey carried out during these two years showed that the damage varied between 10 and 50%. The incidence of the disease

was less in well-drained, sandy loam soils as compared to ill-drained black soils. The disease generally appeared in crop sown during the month of August. Earlier sown crop was almost free from the disease.

An analysis of the meteorological data showed that the disease is favoured by humidity around 90% and temperature around 95°F in the later part of September.

Only three isolates out of a total of sixteen were found to be pathogenic. All the isolates failed to infect any crop other than *guar*. All the three isolates belong to *F. moniliforme* (Sheld.) emend Snyder et Hansen.

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Institute of Agriculture,
Anand.

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PHYLLODY IN SESAMUM (SESAMUM ORIENTALE L.)

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Sesamum (*Sesamum orientale* L.) is an important oilseed crop grown all over India and with an average area of 5.5 million acres under cultivation annually, it forms the chief source of a sweet oil extensively used for domestic purposes. The growers lose a considerable amount of the crop due to phyllody disease which is present in more or less severe form in all the sesamum growing tracts in the country. In the experimental plots of the Indian Agricultural Research Institute, New Delhi, as much as 15 to 50 percent incidence has been recorded during the last three years. Besides *S. orientale*, two other species namely *S. indicatum* (Ramanujam, 1944) and *S. radiatum* Schum. and Thonn. have also been found severely affected with the disease. Almost 100 percent infection in the sesamum crop of 1947 was observed in the district of Raichur (Hyderabad). Robertson (1928) has stated that in certain districts of Burma as much as 80 to 90 percent of the plants may be affected with the disease. The loss in yield is due to the transformation of the floral organs into leafy structures with the result that seed formation does not occur.

The disease has been attributed to different causes by earlier workers. Kashi Ram (1930) considered it a physiological disease induced by early sowings and heavy rainfall. Pal and Pushkarnath (1935) were the first to prove the systemic nature of the disease by grafting and have suggested that it is possibly caused by a virus. Rhind, Odell, and Thet Su (1937) from their studies, however, concluded that phyllody may be due to "a failure of the normal progress of the reproductive phase induced by a combination of environmental conditions acting on complex genetic groupings and that it is a case of the return in varying degrees of the reproductive tissues to the vegetative condition. The possibility of a virus being the agent is not excluded wholly". After the findings of these workers the problem does not appear to have received adequate attention till 1952 when systematic work was undertaken in the Division of Mycology and Plant Pathology of this Institute to establish the nature of the disease and to determine its mode of transmission in nature.

SYMPTOMS OF THE DISEASE

The disease is characterized in the flowering stage when one or more floral parts are transformed into green, leaf-like structures followed by abundant vegetative growth. The calyx which is gamosepalous in normal flowers becomes polysepalous and has multicostate venation. The five sepals become leaf-like but are smaller in size. The corolla may be partially or completely green depending upon the stage of infection. Whereas the normal flowers are zygomorphic, phylloid flowers usually acquire actinomorphic symmetry and the

gamopetalous corolla may become polypetalous. The veins of the sepals and petals in the phylloid flowers are usually thick and prominent. Plate 1-fig. 1 shows different stages of phyllody in the flowers along with healthy flower and capsule.

The stamens generally retain their normal shape but become green in colour. However, in some cases the filament may be flattened to some extent showing its tendency to become leaf-like. The green anthers are indehiscent and do not contain any functional pollen grain. In a normal flower there are only 4 stamens but a phylloid flower bears 5 stamens.

The carpels are transformed into two leafy outgrowths which form a pseudo-syncarpous ovary by their fusion at the margins. This false ovary becomes very much enlarged and flattened. It has a soft texture and a wrinkled surface due to the thickening of the veins of the carpellary wall (Fig. 1-d). In some cases in place of the ovary there may be produced two foliaceous structures fully separated from each other. Inside the ovary instead of the ovules there are small petiole-like outgrowths which later grow and burst through the wall of the false ovary producing small shoots. These shoots continue to grow and produce more leaves and phylloid flowers. Thus it seems as if the flower pedicel has grown through the flower and has an unlimited growth (Fig. 1-e). The normal flowers have very short pedicels but the stalk of a phylloid flower is generally elongated. Vein clearing in different structures is common in the infected plants.

As the size of the foliage leaves in the phylloid shoots is very much reduced and the internodes are shortened, the phylloid flowers and new shoots produced from them are crowded together giving a bunchy-top look to the plant. Plate I-fig. 2 shows naturally infected plants in the field.

Some varieties of sesamum normally develop only one flower in the axil of each leaf and there are present two orange yellow glands, one at each side of the base of the flower stalk. In diseased shoots these glands grow into small phylloid flowers resulting in three such flowers per axil (Fig. 1-g).

It has been observed that the plants may be partially or fully affected. If the disease appears with the first flower, the whole of the plant gets diseased and it does not bear any normal flowers. However, when the infection starts at a later stage, capsules are formed on the lower portions and phylloid flowers are present on the top of the main branches and on the new shoots that are produced from the lower portion. The capsules formed just below the diseased portion, although normal in appearance, contain shrivelled seed which is not viable.

Transmission of the disease

More than 100 plants of sesamum were successfully grafted with the diseased scions under insect proof conditions. The disease was transmitted in all the grafts and the symptoms appeared after a

period of 13 to 45 days depending upon the growth of the plants and weather conditions (Plate II-Fig. 1). Plants grafted in the months of May to October generally showed the disease within 13 to 31 days while those grafted in the remaining months of the year showed the disease symptoms in 32 to 45 days. The disease was also transmitted to *Sesamum occidentale* Heer and Regel by grafting.

Plants raised in the insect proof house from partially affected plant seed appeared to be healthy so that the disease is not seed-borne.

An intensive survey of insects on sesamum revealed that jassids, white-flies, capsids and fulgorids were of common occurrence. However, jassids occurring on the crop received particular attention as some jassids have been reported to be vectors of such virus diseases. In transmission tests insect populations were collected from naturally infected plants in the field, re-fed on diseased sesamum plants for a few days and then liberated on test plants inside the insect proof house for varying periods. Among the insects tried so far *Eutettix phycitis* Dis., *Balclutha* sp., *Empoasca flavescens* Fabr. and the white-fly (*Bemisia tabaci* Genn.) failed to transmit the disease. Positive transmissions were, however, obtained easily in tests with the jassid, *Deltocephalus* sp. The disease was transmitted in 11 out of 13 plants of sesamum. The insects after feeding for 5 to 10 days on diseased plants were liberated on test plants and allowed to feed for 7 to 20 days. Altogether 3 to 9 jassids per plant were released. The symptoms appeared after 33 to 59 days from the date of first feeding on the test plant. The infected plants first showed vein clearing and later produced phylloid flowers subsequently followed by profuse vegetative growth as shown in Plate II-fig. 2. The symptoms were identical with those observed in the naturally affected plants. All these transmission tests with the jassid, *Deltocephalus* sp. were mostly carried out in the months of October and November, 1954 whereas the normal period for the crop is mid-June to mid-October.

Further work is in progress and would be reported later.

The members of the genus *Deltocephalus* Burmeister have been reported to be vectors of disease like winter wheat mosaic and dwarf disease of rice.

The specimens of the jassid (*Deltocephalus* sp.) were identified by the Director, Commonwealth Institute of Entomology, London, to whom grateful thanks are due. Thanks are also due to the Indian Central Oilseeds Committee for financing the scheme under which this work was carried out.

Summary

Phyllody disease of sesamum (*Sesamum orientale* L.) is characterized by the transformation of the floral parts into green leafy structures resulting in complete sterility of the affected plant portions.

The virus nature of the disease has been established and the jassid, *Deltocephalus* sp., has been shown to be the vector of the virus.

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Explanation of Plates

Plate I

Fig. 1. (a). Normal flower. (b). Normal capsule. (c). Partially phylloid flower of sesamum showing the corolla with actinomorphic symmetry. (d). Partially phylloid flower with a false ovary coming out of the green corolla tube with thick and prominent veins on both. (e). Phylloid shoots coming out from the middle of the phylloid flower. (f). A completely phylloid flower with polypetalous corolla. (g). A small phylloid flower.

Fig. 2. Sesamum plants affected with phyllody disease in nature. Note the capsules on the lower portion and phylloid bunches at the top of some branches.

Plate II

Fig. 1. Diseased scion grafted on healthy sesamum stock. The new shoots from the stock show typical symptoms of phyllody.

Fig. 2. Sesamum plant infected through the agency of the viruliferous jassids, (*Deltocephalus* sp.).

PLATE I

Fig. 1—Sesamum phyllody.

Fig. 2—Sesamum phyllody.

PLATE II

Fig. 1—Sesamum phyllody.

Fig. 2—Sesamum phyllody.

BACTERIAL WILT OF EGGPLANT

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Introduction

Eggplant (*Solanum melongena* L.) is an important vegetable crop in West Bengal. It is grown both in the Khariff and Rabi seasons. The plants, grown during Khariff (May to October), suffer from a wilt disease caused by a bacterium. The disease appears in the fields during later part of August when the plants come to flower and begin to bear fruits. Young plants however, are not affected. Wilting which is sudden in nature is characterised by the drooping down of the young top leaves and shoots of the plants. The attacked plants die within 3-5 days of the appearance of the first recognizable symptoms of the disease. Water-soaked areas in the form of black streaks are often noticed on the stem. In severe cases of attack, these areas rot completely and bacteria ooze out in the form of minute drops from these places. The infected region of the stem or root, when cut across, shows brown discolouration in the vascular region. Bacteria come out readily in the form of a milky white stream from the vascular region of the cut end when it is placed in water. The disease is more severe in the years of heavy monsoon when the fields become very frequently water logged, if not flooded. In the years of light monsoon, however, the disease is not so serious. Survey, undertaken in two important egg-plant growing areas to determine the loss caused by the disease has shown that on an average 15 to 23 percent plants die before they bear any fruits and the average reduction in yield may be 54.6 to 62.5 percent due to further death of the bearing plants before full maturity.

Since there is no earlier recorded of bacterial wilt of brinjal from India, a detailed study of the organism isolated was undertaken with a view to identify it. Some preliminary experiments on the survival of the pathogen were also done and the results obtained are reported here.

Isolations :—

The organism was easily isolated by poured plate method and the pure culture was made by streak method. The stock culture was maintained on nutrient-agar. As the organism looses its virulence in artificial culture quickly, it was passed through its host (eggplant), at a periodic interval of 1½-2 months to maintain its vigour. Isolations made from different samples have shown that one particular organism is involved in all the cases.

Morphology :—

The organism is a short rod measuring 3.2-6.4 × 1.6μ, remains in chains in 24 hours old culture, but becomes single later. The organism is gram-negative, non-acid fast, non-capsulated, non-spore forming. The cells are motile by means of polar flagella.

TABLE 1

Growth characters on different media

Medium	Growth characters.
Potato dextrose agar slants.	Spreading, margin entire, surface convex, smooth, glistening, white, opaque in 24 hours' old culture. Colour changed to Dresden brown* with age, of slightly gelatinous consistency. Flourescent green colour appears in 48-72 hours' old cultures, which disappears after 144 hours' growth.
Potato slopes.	Copious, shiny, creamy, spreading over the entire cylinder forming a rugose surface, in 9-10 days the entire cylinder is turned deep brown to light black.
Nutrient broth agar slant.	Growth medium, umbilicate, rough, dull, filiform to echinulate, margin entire to repand, not gelatinous, opaque, pale olive buff. Colour changed to Dresden brown after a week's growth. Green flourescent colour appears in 48-72 hours' old culture which disappears with age. Colouration generally starts from the top and gradually proceeds towards the base.
Nutrient broth.	Growth slightly cloudy, no sediment formation, ring formed in 24 hours' old culture, floccules present at the bottom after 96-120 hours' of growth. Colour changed to light brown after a weeks' growth.
Meat infusion agar slant.	Growth moderate in 35 hours' old culture, surface ridged, white, transparent with faint yellow tinge, with green flourescence.
Meat infusion glucose agar slants.	Growth moderate, surface ridged, margin entire, white with faint yellow tinge and a green flourescent colour in young culture, colour changed to light brown after a week's growth.

* according to Ridgway.

Bio-chemical tests

Acid but no gas is formed in *Dextrose*, *Sucrose*, *Lactose* and *Glycerol*. *Starch*-hydrolysed, *Gelatin*-liquifection slight, M. R. and V. P.—negative, NH_3-positive, NO_2-negative, H_2S-negative. *Indole*-Not produced.

Action on litmus milk—Acid production is noticed within 48-72 hours of growth. In both cream-free as well as full creamed milk, acid is produced. Acid production is followed by coagulation of casein which takes place within 15 to 20 days of growth. Then the liquid is slowly cleared off and the casein is precipitated within 4-5 week's of growth.

Pathogenecity tests

To test whether the organism isolated is pathogenic to the egg-plants and other common plants of Solanaceae, inoculation experiments were carried out on egg-plant (*Solanum melongena* L.), as well as on potato (*Solanum tuberosum* L.), tomato (*Lycopersicum esculentum* L.) and tobacco (*Nicotiana tabacum*). Pathogenecity experiments were conducted in the winter season. The minimum temperature during the period varied from 50-60°F and the maximum temperature 80°-90°F.

Ten seedlings of egg plant (3 weeks old) raised from sterilized seed in sterilized soil were transplanted each into a 250 c.c. erlenmeyer flask containing 100 c. c. of the sterile nutrient solution (Prepared from *Manurin* tablets supplied by Messers. India Alkalies Ltd.). In each flask 5 c.c. of the bacterial culture in nutrient broth grown at 35°C for 24 hours were added as inoculum. The flasks were wrapped with black paper to prevent algal growth. Nutrient solution and sterile distilled water were periodically added to replenish the loss of water and nutrients. A similar set of ten flasks was used as controls in which no inoculum was added. To facilitate the attack, in each plant a few fine rootlets were cut by fine scissors Plants growing in inoculated flasks showed characteristic symptoms of wilting within 10 days and all of them completely wilted within the next 4-5 days, while the controls remained perfectly healthy. The wilted plants showed poor development of root system. The same organism could be reisolated from the wilted plants. This experiment was repeated with sterilized soil-extract solution instead of sterile nutrient solution and the same results were obtained.

Pathogenecity tests were also conducted in earthen pots containing sterilized soil (with formalin) and later on inoculated with 6 days' old bacterial cultures grown on sterilized potato slopes. Fifteen pots were taken and in each one seedling (3 weeks' old) was transplanted after 7 days' of soil inoculation. A similar set of ten plants grown in sterilized but uninoculated soil was kept as control. It was observed that even after a month's growth, no sign of wilting was noticed in the inoculated sets. In 10 pots, the soil was carefully removed and the

fine roots were punctured by scissors The plants in these pots showed typical symptoms of wilting within 10 days of puncture, whereas of the 5 plants, with uninjured roots, only one showed wilting after a period of two months.

Pathogenecity experiments were also conducted on Tomato, Tobacco and Potato. Tomato seedlings were grown in nutrient solution whereas Tobacco and Potato were grown in sterilized soil. In all the cases, a set of ten plants (three weeks' old) was taken while a similar set was kept as control.

In cases of Tobacco and Potato, inoculations were made with bacterial suspension by wounding the stem in the basal region near soil level whereas in Tomato bacterial suspension was added to the nutrient solution and roots were slightly injured to facilitate the attack.

In all the cases symptoms of wilting were noticed within 2-3 weeks after the inoculation. The top leaves showed signs of drooping, shrivelling and wilting. Symptoms gradually proceed towards the base of the stem where discolouration and shrivelling could be noticed. All the inoculated plants collasped within a week after the appearance of the disease. The same organism could be reisolated in all the cases. The inoculation experiment on tomato was repeated on pots with similar findings. All the experiments were repeated twice to get confirmatory results.

The foregoing experiments conclusively showed that the organism is pathogenic to eggplant and it can also infect other Solanaceous crops like tobacco, tomato and potato.

Identification of the organism

The morphological and cultural characters place the organism under the genus *Pseudomonas*. The host-range, the symptoms produced and the biochemical activities correspond with those of *Pseudomonas solanacearum* (Smith) Smith. From its reaction on litmus milk, namely production of acid, the organism is identified as *Pseudomonas solanacearum* var. asiaticum (Smith) Stapp according to Dowson (1949).

Pseudomonas solanacearum var. asiaticum was described by Smith (1914) and later by Maia and D'Oliveira (1945) from Portugal on potato and tomato. As far as it is known, *Pseudomonas solanacearum* var. asiaticum has not yet been recorded on eggplant. *Pseudomonas solanacearum* var. asiaticum causing wilt of potato, has however been recently reported from this laboratory (Mukherjee and Chattopadhyay, 1955).

Survival of the pathogen

Pseudomonas solanacearum var. asiaticum is a soil inhabitant organism. From the standpoint of survival of the organism and its pathogenesis, two factors may be considered important, (a) capacity of the bacteria to remain alive in the soil in the absence of host plant

and (b) in the dead plant remains. Accordingly preliminary experiperiments were undertaken in these directions.

(a) *Survival in soil:* To find out how long the organism *Pseudomonas solanacearum* var. asiaticum can survive in the soil in the absence of host plant, a set of six flasks each containing 25 gm. of soil was sterilized in autoclave at 20 lbs pressure for 30 minutes and later on inoculated with 10 c. c. of 24 hours old bacterial culture. At monthly interval, a very small quantity of the sample was taken out and was examined to find out whether the organism was still in a living condition. The experiment was started in the last week of July, 1953 and the organism was found to be alive even upto last week of November, 1954 i. e. upto a period of 16 months. The pH of the soil at the end of the experiment at the last week of December, 1954 was 7.5 (as compared with 7.0 in the initial stage) and moisture 1.71% as compared with 6% normal moisture in the soil of the sample used.

This proves that the organism though a non-spore-former can remain alive for quite a long period even under conditions of dessication.

(b) *Survival in the diseased plant remains:* Samples of wilted egg-plants were collected in the month of September, 1953. They were cut into small bits of 1½-2″, made airdry and were kept in a dry condition in well stoppered glass jar, at room temperature. Isolations made at monthly intervals showed that the organism was alive till the month of July, 1954. This finding shows the possibility of the disease being carried over from the harvesting of one season to the beginning of the next season in the dried dead plant remains. Besides soil, the diseased plant remains may also constitute an important source of infection.

Summary

1. Egg-plants grown in West Bengal during the months of May to October suffer from a wilt disease caused by a bacterium.

2. The disease appears in the field when the plants come to flower. It is very serious and destructive particularly in years of heavy rainfall.

3. The organism is identified as *Pseudomonas solanacearum* var. asiaticum (Smith) Stapp. As far as it is known, this organism is being reported for the first time on egg-plant.

4. *Pseudomonas solanacearum* var. asiaticum besides infecting eggplant, can also produce wilting on potato, tomato and tobacco under artificial conditions.

5. *Pseudomonas solanacearum* var. asiaticum can remain alive in the soil (under laboratory condition) upto a period of 16 months. It can also remain alive in the diseased plant remains upto a period of 9 months.

Sincere thanks are due to Dr. W. J. Dowson, of Botany School, Cambridge for his help in the identification of the organism.

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TWO NEW XANTHOMONAS SPECIES ON LEGUMES

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Introduction

Two bacterial diseases of leguminous plants viz. *Butea frondosa* Konig. and *Tephrosia purpurea* Pers. were observed at different places in Bombay State during the rainy season of 1953. When these were studied for morphological, cultural and biochemical characters, it was observed that they were indistinguishable from other *Xanthomonas* species but were highly specific to their own suscepts, the latter being a unique character of the genus *Xanthomonas* as observed by Wernham (1948), Patel, Dhande and Kulkarni (1951) and Patel and Bhatt (1954). They were, therefore, designated as novum species. This paper is a more detailed account of a short note published by Patel and Bhatt (1954).

Suscepts

Butea frondosa, sacred to *Soma* (Moon) and one of the most beautiful trees of the plains of India has its every part of some use. It yields gum from the bark, oil solely of medicinal value from the seeds, dye and tan from the flowers, strong fibre for paper making and cordage, lac of the second best quality and the leaves are used as fodder for the buffaloes and the elephants and as a valuable manure. The most economical use of the leaves in India is seen when dishes are prepared from them to serve food. A severe bacterial disease was found on the leaves and the petiole at Ambarnath (Thana) in September, 1953.

Tephrosia purpurea, a legume, is much relished by goats and sheep as fodder. It is also made use of in soil conservation. The bacterial disease on it was first noticed by the senior author on a few isolated plants at Agra in November, 1952. However, the disease was found quite prevalent on Agricultural College Farm, Poona in August, 1953 and isolates from the Agra and Poona materials were found to be quite similar in all the subsequent tests.

Isolation and Inoculation

The pathogens from each were isolated separately by usual poured plate method. In each case, small, raised, circular, shining, yellow colonies appeared after 48 hours' incubation at 30°C. and a single colony was subcultured and subsequently maintained on neutral potato dextrose agar slants.

Inoculation experiments were carried out to test the pathogenicity of the isolates under study on their respective hosts. A separate atomiser and a moist chamber were kept for each pathogen and its

host. Both the cultures produced disease symptoms on their respective hosts within 15 days and a single colony reisolated proved identical to previous isolates in all the subsequent tests.

Symptoms

The symptoms described below are the same as those observed in nature and on plants artificially inoculated.

The pathogen on *Butea frondosa* produces in early stage of the disease, a few to numerous, minute, water-soaked areas with brown centre and pale yellow halo, measuring 0.3—0.5 mm., fairly well distributed all over the leaf surface (Fig. 1). Young leaves, injured tender petiole and stem are easily infected under high humidity and continuous rains so essential for infection. With the progress of the disease, the spots increase in size to 0.8—1.2 mm., become round to angular and dark brown to jet black. At Ambarnath where rain exceeds 100 inches and humid weather with intermittent heavy showers prevails throughout the rainy season, the bacterial infection on young leaves is sometimes so severe that even a pin point area is not left over without infection. Such leaves dry and shed off before maturity. It also produces canker on injured tender petiole (Fig. 2).

The pathogen on *Tephrosia purpurea* produces a few, small, round, water-soaked areas measuring initially 0.5—0.7 mm. In the beginning, the spots are pale brown with a small yellow halo around them which later increase in size to 1-2 mm., and become dark brown to jet black (Fig. 3). The pathogen infects injured tender stem and rachis besides leaves.

Morphology and Cultural Characters

The pathogens from *B. frondosa* and *T. purpurea* designated as *Xanthomonas buteae* and *X. tephrosiae* respectively were found to be short rods, 0.5×2.0 μ in size, Gram negative, capsulated, non-spore former and motile by single polar flagellum Agar colonies were circular, entire, smooth, raised, and butyrous. *X. buteae* produced sulfur yellow (R) and amber yellow (R) on potato dextrose and nutrient dextrose respectively while *X. tephrosiae* produced martius yellow (R) and picric yellow (R) respectively on both the media. They made good growth on yeast-dextrose-chalk and Krum weide's agars. *X. buteae* made no growth in synthetic nitrate and Czapek's media while *X. tephrosiae* made good growth in both the media. Optimum temperature for growth was 27° – 30°C. and thermal death point 51°C.

Biochemical Reactions

Unless otherwise mentioned, the following observations are for both the cultures. All the media and their corresponding tests were carried out according to the methods recommended in the Manual (Soc. of American Bacteriologists 1945) and the readings recorded after 8 days at 30°C.

Gelatin liquefied, starch hydrolysed and casein digested. Litmus milk was changed from mauve (R) to roseline pink (R) in 4-5 days and peptonised completely in 8-10 days leaving a transparent liquid at the top. Indol was not produced from tryptophane. Hydrogen sulphide and ammonia produced from peptone. *X. buteae* completely liquefied Loeffler's blood serum in 12 days while *X. tephrosiae* did so fairly in 16 days. Both were M. R. and V. P. negative. None could grow in Cohn's ammonium tartarate and Clara's asparagine media but grew well in Uschinsky's glycerol ammonium lactate medium with alkaline reaction. *X. buteae* made excellent growth with acid reaction in Fermi and Montesano's glycerol ammonium tartarate medium while *X. tephrosiae* made slight growth with no change in pH of the medium. Nitrite and ammonia were not produced from nitrate. Sodium chloride concentration tolerated upto 3 per cent and pH from 4.0 to 9.5 with optimum growth at 7 pH.

Utilisation of Carbon Compounds

When fermentation study with 1 per cent carbon compounds or 0.1 per cent organic acids incorporated in a synthetic medium, neutralised and sterilised in Arnold for 3 successive days was carried out, good growth and acid without gas were produced from arabinose, galactose, levulose, rhamnose, xylose, dextrose, lactose, maltose, sucrose, starch and alkaline reaction in acetic, citric and lactic acids but no growth in salicin, cellulose and acids like benzoic, formic, oxalic, salycilic, tannic and tartaric. *X. buteae* made good growth with acidity in glycerol and mannitol but made fair growth with slight acidity in dulcitol and raffinose while *X. tephrosiae* made good growth with acid reaction in mannitol and raffinose and slight growth with slight acidity in dulcitol and glycerol.

Utilisation of Nitrogenous Compounds

Utilisation of 0.14 per cent of nitrogen from inorganic and 0.1 per cent of organic nitrogenous compounds added separately to modified Richards' medium as suggested by Patel and Kulkarni (1949) was studied in liquid medium. When 3 loops of each culture allowed to grow for 48 hours in peptone dextrose broth were inoculated and growth recorded after 4 serial transplants, a method followed by Starr and Weiss (1945), it was observed that ammonium citrate, ammonium di-hydrogen phosphate, ammonium nitrate, ammonium oxalate, ammonium sulphate, ammonium tartarate, alanine, asparagine, aspartic acid, creatin, creatinine, cystine, guanidine hydrochloride, glycine, glucocyamine, leucine, methionine, norvaline, ornithine hydrobromide, phenyl alanine, serine, threonine, tryptophane, tyrosine, urea and valine supported the growth while nitrates of Ag, Ba, Ca, Cd, Cu and Zn along with nitrite of sodium proved olygodynamic or toxic ; glutamic acid and proteose peptone supported the growth of the organisms even when carbon was not supplied in the experiment on organic nitrogenous compounds. Nitrogen from nitrates of magnesium, potassium and sodium was not utilised by *X. buteae* but *X. tephrosiae* utilised it and made good growth in all.

Host Range

In order to avoid confusion regarding the character of host specificity, it is a common practice in this laboratory to inoculate all the available hosts on which a bacterial disease is described previously from this place. Whenever a pathogen under study is found to infect any one or more of the previously described host or hosts, cross inoculations of such cultures on the hosts concerned are made to see that the recent diseased host is not a collateral one. Patel and Bhatt (1954) have shown that 60 per cent of the *Xanthomonas* species described from India parasitise the members of Leguminosae only. It was, therefore, thought worthwhile to inoculate as many related and unrelated hosts as possible to confirm additional hosts for these two cultures.

When *Acacia arabica* Willd., *A. catechu* Willd., *A. decurrens* Willd., *A. melanoxylon* Br., *Aegle marmelos* Corr., *Alysicarpus bupleurifolius* DC., *A. hamosus* Edgew.. *A. longifolius* W. & A., *A. monilifer* DC., *A. pubescens* Law., *A. rugosus* DC., *A. tetragonolobus* Edgew., *A vaginalis* DC., *Amaranthus viridis* L., *Arachis hypogaea* L., *Begonia* sp., *Brassica oleracea* L., *Bridelia hamiltoniana* Wall., *Butea frondosa* Konig., *Caesalpinia pulcherrima* Swartz., *C. sepiaria* Roxb., *Capsicum annuum* L., *Cajanus cajan* Millsp., *Cassia alata* L., *C. didymobotrya* Fraesen, *C. hirsuta* L., *C. siamea* Lam., *C. tora* L., *Centrosema pubescens* L., *Cicer arietinum* L., *Citrus aurantifolia* Sw., *Clerodendron phlomoides* L., *Crotolaria anagyroides* H. B. & K., *C. juncea* L., *C. striata* DC., *Cyamopsis tetragonoloba* (L.) Taub., *Desmodium diffusum* DC., *D. gangeticum* DC., *Dolichos biflorus* L., *D. lablab* L., *Erythrina indica* Lam., *Euphorbia pulcherrima* Willd., *Gossypium herbaceum* L., *Indigofera argenta* L., *I. arrecta* Benth., *I. glandulosa* Willd., *I. tinctoria* L., *Ipomoea muricata* R. & Sch., *Lathyrus odoratus* L., *L. sativus* L., *Lawsonia alba* Lam., *Leucaena glauca* L., *Lochnera pusilla* K. Schum., *Medicago sativa* L., *Melilotus indica* All., *Moringa pterigosperma* Gaertn., *Phaseolus aconitifolius* Jacq., *P. angularis* Wight, *P. coccineus* Lam., *P. lunatus* L., *P. mungo* var. *radiatus* L., *P. vulgaris* L., *Piper betle* L., *Pisum arvense* L., *P. sativum* L., *Poinciana regia* Bojer., *Pueraria phaseoloides* Borth., *Ricinus communis* L., *Sesbania aculeata* Poir., *S. aegyptiaca* Prain., *Soja max* (L.) Piper, *Stizolobium deeringianum* Bort, *Tamarindus indica* L., *Tectona grandis* L., *Tephrosia candida* DC., *T. purpurea* Pers., *Trichodesma zeylanicum* R. & Br., *Trifolium alexandriacum* L., *Trigonella foenum-graecum* L., *Vigna catjang* Walp., and *Xanthium strumarium* L., were inoculated in a separate series with a 48 hour culture of *X. buteae* and *X. tephrosiae*, it was observed that *X. buteae* infected only *Butea frondosa* and *X. tephrosiae* only *Tephrosia purpurea.*

Taxonomy and Nomenclature

The morphological, cultural and biochemical characters of these two cultures are quite similar to those of the genus *Xanthomonas* when referred to Dowson (1939) and thus it is beyond doubt that they belong to the genus *Xanthomonas.* As mentioned pre-

On leaves of *Mimusops hexandra*, Patna, Bihar, 8-10-1952, leg. A. S. Yadav.

4. *Olivea colebrookiana* (Barclay) Thirumal. & Yadav comb. nov.

Syn. *Uredo colebrookiana* Barcl. in Jour. Asiatic Soc. Bengal, LX, 227 p. 1891.

Uredia hypophyllous, developing on pale-yellow infection spots, densely grouped and often covering the entire leaf surface, golden yellow, subepidermal, erumpent and pulverulent. Urediosori formed by strands of hyphae emerging through and distending stomata, forming sorus above level of epidermis, covered marginally by basket of incurved paraphyses; urediospores golden-yellow, ovate to spherical, 14-26×12-16μ., epispore minutely aculeate. Telia similar to uredia in sorus structure, pale-yellow in fresh material, appearing white in exsiccati material; teliospores sessile, obclavate to cylindric, with orange-yellow contents in fresh material, thin-walled, hyaline, 32-48×8-12μ., germinating at maturity by the prolongation of spore apex, promycelium external, four-celled. Sporidia globular, thin-walled, up to 8-10μ. in diameter.

Hab. on leaves of *Colebrookia oppositifolia*, Parasnath, Bihar, 11-4-1954, leg. A.S. Yadav. (Figs. 6 and 7).

Olivea colebrookiana till now known only in the uredial stage, parasitises the leaves of *Colebrookia oppositifolia* a member of the Labiatae. The rust is widely distributed in India, having been collected by one of the authors (Thirumalachar) in Simla (type locality for *U. colebrookiana*), Bhowali (U.P.), Mercara (Coorg) and Khandala (Bombay). While the rust is known so far only in the uredial stage, collections made at Parasnath during the months of March and April showed abundant development of telia which were macroscopically indistinguishable from the uredia. The telia under magnification with a field lens appeared compact and crust-like. A similar condition has been observed in the case of *Olivea tectonae* (Racib.) Thirumal. (Thirumalachar Curr. Sci. 18 : 175-177). The teliospores germinate immediately in the sorus developing promycelia and sporidia.

5. *Cerotelium bauhiniae* Thirumal. & Yadav. sp. nov.

Uredia hypophyllous, minute, pale brown, subepidermal, erumpent, lined with hyphoid paraphyses. Uredispores sessile, obovate to ellipsoid, yellow-brown, with indistinct germ pores, 20-28× 20-24μ., epispore minutely echinulate. Telia minute, gregarious, associated with uredia, subepidermal, appearing as crusts in early stages, later becoming erumpent; teliospores occurring in chains, showing lateral coalescence, forming a column at the base and becoming separated and pulverulent at the apex, 4 to 6 spores in a chain, angularly globoid to cuboid, thin-walled, smooth, 10-12×9-12μ., germinating at maturity.

Hab. on leaves of *Bauhinia anguinia*, Parasnath, 27-2-1953, leg. A.S. Yadav (Type). on *B. vahlii*, Parasnath, 27-2-1953, leg. A. S. Yadav. (Figs. 8 to 10).

Indian Phytopathology 8(2):140-145,1955.

viously, the genus *Xanthomonas* is unique in the matter of ho specificity and new species are being created on such a charact only. Even though hosts of similar to distinctly dissimilar flo structure are tried, the pathogens are found restricted to their ov suscepts from which they are originally isolated. So far, there is record of a *Xanthomonas* species on either of the hosts when referr to Elliott (1951) and as the pathogens are highly specific to th suscepts, it is thought reasonable to assign each a status of nov species.

Xanthomonas buteae Bhatt, Patel and Thirumalachar sp. no On leaves of *Butea frondosa* Konig. Found at Ambarnath (Thar in September, 1953. Leg. V. V. Bhatt.

Short rods ; single polar flagellum ; Gram negative ; capsulated non-spore former ; agar colonies, smooth, circular, raised, butyro and yellow ; gelatin liquefied ; starch strongly hydrolysed ; case digested ; milk peptonised and litmus reduced ; hydrogen sulphi and ammonia produced from peptone ; nitrite not produced fro nitrate ; no growth in synthetic nitrate and Czapek's media ; ac without gas from arabinose, dextrose, lactose, sucrose, and starch no growth in salicin ; optimum temperature for growth 27°-30 C thermal death point about 51°C.

Xanthomonas tephrosiae Bhatt, Patel and Thirumalachar s nov. On leaves of *Tephrosia purpurea* Pers. Found at Agra an Agricultural College Farm, Poona in August, 1953. Leg. V. V. Bhat

Short rods ; single polar flagellum ; Gram negative ; capsulated non-spore former ; agar colonies, smooth, circular, raised, butyro and yellow ; gelatin liquefied ; starch hydrolvsed ; casein digested milk peptonised and litmus reduced ; hydrogen sulphide and ammon produced from peptone ; nitrite and ammonia not produced fro nitrate ; good growth in synthetic nitrate and Czapek's media ; ac without gas from arabinose, dextrose, lactose, sucrose and starch ; n growth in salicin ; optimum temperature for growth 27°-30°C thermal death point about 51°C.

SUMMARY

Two bacterial diseases of leguminous plants viz. *Butea frondo* and *Tephrosia purpurea* are described with symptomatical, morph logical, cultural, biochemical and hosts responses of the pathoger From these studies, they are found to belong to the genus *Xanth monas* and have been alloted a status of new species on account their host specificity.

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EXPLANATION OF FIGURES

g. 1. Water-soaked areas with brown centre and yellow halo on the young leaves of *Butea frondosa*.

g. 2. Canker production on the injured petiole of *Butea frondosa*, (a) Healthy (b) Diseased.

g. 3. A few, round, dark brown to jet black spots with small pale yellow halo on the leaves of *Tephrosia purpurea*,

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Uredia minuta, hypophylla, pallide brunnea, subepidermalia, erumpentia, hyphoides paraphysibus intus ad muros dispositis. Uredio-sporae sessiles obovatae vel ellipsoideae, luteo-brunneae, poris germinationis indistinctae, 20-28×20-24μ.. episporio echinulatae, Telia minuta, gregaria, urediis associata, subepidermalia, initio crustata, postea erumpentia; teliosporae catenatae, lateraliter coalescentes, columnam in basi efformantes, in apice separatae atque pulverulentae, sporae 4-6 in singulis catenis, angulariter globoideae vel cuboideae, hyalinae, tenuiter parietatae, leves, magnitudinis 10-12×9-12μ.. maturitatae germinantes,

The presence of hyphoid paraphyses in the uredia first indicated that the rust may be a species of *Cerotelium*, and a careful search of numerous infected leaves revealed the presence of telia which are minute and sparsely distributed. While only *Uromyces vestergreni* Syd. is known on species of *Bauhinia* in India, several other species of *Uromyces* and *Scopella* have been reported on the host genus *Bauhinia* in other countries. No species of *Cerotelium* has been described on *Bauhinia*.

6. *Cerotelium kirganeliae* Thirumal. & Yadav. sp. nov.

Infection spots pale yellow. Pycnia, aecia and uredia unknown. Telia hypophyllous, white, subepidermal, erumpent and pulverulent. Teliospores one-celled, cuboid to polygonal, hyaline, thin-walled, developed in chains in basipetal succession ; telial chains laterally coalescent, teliospores pulverulent at the apex, separating away at the apex, germinating immediately at maturity, 10-12×8-12μ.

Hab. on leaves of *Kirganelia reticulata*, Sabour Agricultural College, Bhagalpur, 1-1-1953, leg. M.J. Thirumalachar. (Figs. 11 and 12).

Infection is maculis pallide lutea. Pycnia, aecia atque uredia ignota. Telia hypophylla, albida, subepidermalia, erumpentes atque pulverulenta. Teliosporae, cuboideae vel angulariter globoideae, catenatae, lateraliter coalescentes, in apice separatae, atque in maturitatae germinantes, hyalinae, tenuiter parietatae, 10-12×8-12μ.

The rust is placed under *Cerotelium* on account of the catenulate teliospores which are coalescent at the base and pulverulent at the apex. The type of uredia offers a very characteristic diagnosis in the genus *Cerotelium* and on account of its absence in the present rust the identification is only tentative.

7. *Uromyces anthyllidis* (Grev.) Schroet.

On leaves of *Trigonella foenum-graecam*, Patna, 8-8-1953, leg. A. S. Yadav.

8. *Uromyces blainvilliae* Berk.

On leaves of *Blainvilliae rhomboidea* (*B. latifolia*), Parasnath, 11-4-1954, leg. A. S. Yadav.

9. *Uromyces hobsoni* vize.

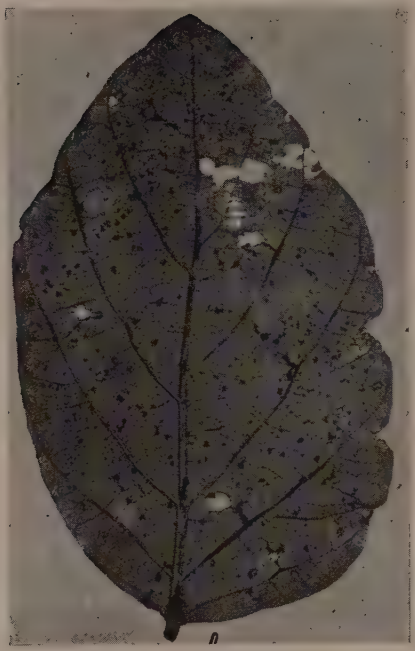

Fig. 1—*Butea frondosa*

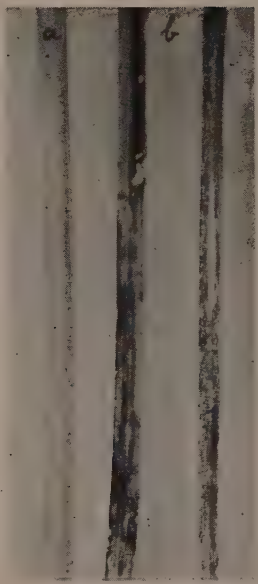

Fig. 2—*Butea frondosa*

Fig. 3—*Tephrosia purpurea*

CONTRIBUTION TO THE KNOWLEDGE OF UREDINEAE OF BIHAR-II*

A. S. YADAV AND M. J. THIRUMALACHAR.

(Accepted for publication, October 12, 1955)

Since the publication of the previous paper on some of the rust fungi in Bihar, further collections have been made and studied. Some of the rust species which are either new to Science or new records for Bihar, are being reported in this paper. The types of the new species have been deposited in the Herb. Crypt. Ind. Orient. New Delhi, Herb. C.M.I., Kew, England, and in the Mycological division, U.S.D.A., Beltsville, Maryland, U.S.A.

1. *Acervulopsora ichnocarpi* (Barcl.) Thirumal. Mycologia 37 : 229, 1945. On leaves of *Ichnocarpus frutescens*, Parasnath, Bihar, 11-4-1954, leg. A.S. Yadav. Abundant production of telia was observed in the material, appearing as white powdery crusts.

2. *Maravalia milletiae* Yadav & Thirumal. sp. nov.

Pycnia and aecia unknown. Uredia hypophyllous on yellowish-brown infection spots, subepidermal, erumpent and pulverulent, yellow, paraphysate, paraphyses clavate and interspersed. Urediospores subglobose to spherical, pale yellowish-brown, pedicellate, measuring 16-20×12-20μ., minutely echinulate, with indistinct germ pores. Telia minute, developed as waxy crusts, pale yellow, subepidermal, erumpent ; teliospores clavate to cylindric, one-celled, pedicellate with orange-yellow contents 32-44×12-14μ., wall hyaline, thin ; spores germinating intrasorum at maturity by the prolongation of spore apex ; promycelium external, four-celled. Pedicel hyaline, 76-88×8-10μ.

Hab. on leaves of *Milletia auriculata*, Parasnath, Bihar, 14-12-1942, leg. A.S. Yadav. (Figs. 1 to 5).

Pycnia atque aecia ignota. Uredia hypophylla, maculis flavo-brunnea, subepidermalia, lutea, erumpentes atque pulverulenta, paraphysata, paraphysibus clavata atque interspersa. Urediosporae subglobosae vel sphaericae, pallide flavo-brunneae, pedicellatae, 16-20×12-20μ., minuteque echinulatae, germinationis poro indistincto. Telia minuta, evolventia ut crustae cereae, pallide lutea, subepidermalia, atque erumpentia ; teliosporae hyalinae, pedicellatae, 32-44× 12-14μ. maturae sporae germinantes in soro, producto sporarum apice, promycelium externum, 4-cellulatum. Pediculi hyalini, 76-88×8-10μ.

3. *Scopella gentilis* (Syd.) Mundkur & Thirumal. in Mycol. pap. No. 40, C.M.I., Kew, England, p. 12-13, 1951.

* Part-I in Indian Phytopathology 6 : 86-91, 1953.

viously, the genus *Xanthomonas* is unique in the matter of host specificity and new species are being created on such a character only. Even though hosts of similar to distinctly dissimilar floral structure are tried, the pathogens are found restricted to their own suscepts from which they are originally isolated. So far, there is no record of a *Xanthomonas* species on either of the hosts when referred to Elliott (1951) and as the pathogens are highly specific to their suscepts, it is thought reasonable to assign each a status of *novum* species.

Xanthomonas buteae Bhatt, Patel and Thirumalachar sp. nov. On leaves of *Butea frondosa* Konig. Found at Ambarnath (Thana) in September, 1953. Leg. V. V. Bhatt.

Short rods ; single polar flagellum ; Gram negative ; capsulated ; non-spore former ; agar colonies, smooth, circular, raised, butyrous and yellow ; gelatin liquefied ; starch strongly hydrolysed ; casein digested ; milk peptonised and litmus reduced ; hydrogen sulphide and ammonia produced from peptone ; nitrite not produced from nitrate ; no growth in synthetic nitrate and Czapek's media ; acid without gas from arabinose, dextrose, lactose, sucrose, and starch ; no growth in salicin ; optimum temperature for growth 27°–30°C.; thermal death point about 51°C.

Xanthomonas tephrosiae Bhatt, Patel and Thirumalachar sp. nov. On leaves of *Tephrosia purpurea* Pers. Found at Agra and Agricultural College Farm, Poona in August, 1953. Leg. V. V. Bhatt.

Short rods ; single polar flagellum ; Gram negative ; capsulated ; non-spore former ; agar colonies, smooth, circular, raised, butyrous and yellow ; gelatin liquefied ; starch hydrolysed ; casein digested ; milk peptonised and litmus reduced ; hydrogen sulphide and ammonia produced from peptone ; nitrite and ammonia not produced from nitrate ; good growth in synthetic nitrate and Czapek's media ; acid without gas from arabinose, dextrose, lactose, sucrose and starch ; no growth in salicin ; optimum temperature for growth 27°–30°C.; thermal death point about 51°C.

SUMMARY

Two bacterial diseases of leguminous plants viz. *Butea frondosa* and *Tephrosia purpurea* are described with symptomatical, morphological, cultural, biochemical and hosts responses of the pathogens. From these studies, they are found to belong to the genus *Xanthomonas* and have been alloted a status of new species on account of their host specificity.

Plant Pathological Lab.,
College of Agriculture,
Poona 5.

On leaves of *Mimusops hexandra*, Patna, Bihar, 8-10-1952, le A. S. Yadav.

4. *Olivea colebrookiana* (Barclay) Thirumal. & Yadav comb. no

Syn. *Uredo colebrookiana* Barcl. in Jour. Asiatic Soc. Beng LX, 227 p. 1891.

Uredia hypophyllous, developing on pale-yellow infection spot densely grouped and often covering the entire leaf surface, golde yellow, subepidermal, erumpent and pulverulent. Urediosori forme by strands of hyphae emerging through and distending stomat forming sorus above level of epidermis, covered marginally by bask of incurved paraphyses; urediospores golden-yellow, ovate to spherica 14-26×12-16μ., epispore minutely aculeate. Telia similar to uredia sorus structure, pale-yellow in fresh material, appearing white exsiccati material; teliospores sessile, obclavate to cylindric, wit orange-yellow contents in fresh material, thin-walled, hyalin 32-48×8-12μ., germinating at maturity by the prolongation of spo apex, promycelium external, four-celled. Sporidia globular, thi walled, up to 8-10μ. in diameter.

Hab. on leaves of *Colebrookia oppositifolia*, Parasnath, Biha 11-4-1954, leg. A.S. Yadav. (Figs. 6 and 7).

Olivea colebrookiana till now known only in the uredial stag parasitises the leaves of *Colebrookia oppositifolia* a member of th Labiatae. The rust is widely distributed in India, having bee collected by one of the authors (Thirumalachar) in Simla (typ locality for *U. colebrookiana*), Bhowali (U.P.), Mercara (Coorg) an Khandala (Bombay). While the rust is known so far only in th uredial stage, collections made at Parasnath during the months March and April showed abundant development of telia which wer macroscopically indistinguishable from the uredia. The telia und magnification with a field lens appeared compact and crust-like. similar condition has been observed in the case of *Olivea tectone* (Racib.) Thirumal. (Thirumalachar Curr. Sci. 18: 175-177). Th teliospores germinate immediately in the sorus developing promycel and sporidia.

5. *Cerotelium bauhiniae* Thirumal. & Yadav. sp. nov.

Uredia hypophyllous, minute, pale brown, subepiderma erumpent, lined with hyphoid paraphyses. Uredispores sessile, ob vate to ellipsoid, yellow-brown, with indistinct germ pores, 20-28 20-24μ., epispore minutely echinulate. Telia minute, gregariou associated with uredia, subepidermal, appearing as crusts in ear stages, later becoming erumpent; teliospores occurring in chain showing lateral coalescence, forming a column at the base and b coming separated and pulverulent at the apex, 4 to 6 spores in chain, angularly globoid to cuboid, thin-walled, smooth, 10-12×9-12μ germinating at maturity.

Hab. on leaves of *Bauhinia anguinia*, Parasnath, 27-2-1953, le A.S. Yadav (Type). on *B. vahlii*, Parasnath, 27-2-1953, leg. A. S. Yada (Figs. 8 to 10).

Uredia minuta, hypophylla, pallide brunnea, subepidermalia, erumpentia, hyphoides paraphysibus intus ad muros dispositis. Uredio-sporae sessiles obovatae vel ellipsoideae, luteo-brunneae, poris germinationis indistinctae, 20-28×20-24μ.. episporio echinulatae, Telia minuta, gregaria, urediis associata, subepidermalia, initio crustata, postea erumpentia; teliosporae catenatae, lateraliter coalescentes, columnam in basi efformantes, in apice separatae atque pulverulentae, sporae 4-6 in singulis catenis, angulariter globoideae vel cuboideae, hyalinae, tenuiter parietatae, leves, magnitudinis 10-12×9-12μ.. maturitatae germinantes,

The presence of hyphoid paraphyses in the uredia first indicated that the rust may be a species of *Cerotelium*, and a careful search of numerous infected leaves revealed the presence of telia which are minute and sparsely distributed. While only *Uromyces vestergreni* Syd. is known on species of *Bauhinia* in India, several other species of *Uromyces* and *Scopella* have been reported on the host genus *Bauhinia* in other countries. No species of *Cerotelium* has been described on *Bauhinia*.

6. *Cerotelium kirganeliae* Thirumal. & Yadav. sp. nov.

Infection spots pale yellow. Pycnia, aecia and uredia unknown. Telia hypophyllous, white, subepidermal, erumpent and pulverulent. Teliospores one-celled, cuboid to polygonal, hyaline, thin-walled, developed in chains in basipetal succession ; telial chains laterally coalescent, teliospores pulverulent at the apex, separating away at the apex, germinating immediately at maturity, 10-12×8-12μ.

Hab. on leaves of *Kirganelia reticulata*, Sabour Agricultural College, Bhagalpur, 1-1-1953, leg. M.J. Thirumalachar. (Figs. 11 and 12).

Infection is maculis pallide lutea. Pycnia, aecia atque uredia ignota. Telia hypophylla, albida, subepidermalia, erumpentes atque pulverulenta. Teliosporae, cuboideae vel angulariter globoideae, catenatae, lateraliter coalescentes, in apice separatae, atque in maturitatae germinantes, hyalinae, tenuiter parietatae, 10-12×8-12μ.

The rust is placed under *Cerotelium* on account of the catenulate teliospores which are coalescent at the base and pulverulent at the apex. The type of uredia offers a very characteristic diagnosis in the genus *Cerotelium* and on account of its absence in the present rust the identification is only tentative.

7. *Uromyces anthyllidis* (Grev.) Schroet.

On leaves of *Trigonella foenum-graecam*, Patna, 8-8-1953, leg. A. S. Yadav.

8. *Uromyces blainvilliae* Berk.

On leaves of *Blainvilliae rhomboidea* (*B. latifolia*), Parasnath, 11-4-1954, leg. A. S. Yadav.

9. *Uromyces hobsoni* vize.

TURN OVER E.

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Explanation of Figures

Fig. 1. Water-soaked areas with brown centre and yellow halo on the young leaves of *Butea frondosa*.

Fig. 2. Canker production on the injured petiole of *Butea frondosa*, (a) Healthy (b) Diseased.

Fig. 3. A few, round, dark brown to jet black spots with small pale yellow halo on the leaves of *Tephrosia purpurea*.

On leaves and stems of *Jasminum scandens*, Parasnath, 11-4-1954, leg. A. S. Yadav.

10. *Uromyces mucunae* Rabenh.

On leaves of *Mucuna pruriens*, Patna, 8-8-1953, leg. A. S. Yadav.

11. *Masseeella capparidis* (Hobson) Dietel in Ber. deutsch. bot. Ges. 12: 332, 1895.

Hab. On leaves of *Flueggea virosa*, Abhaipur hills, 19-9-1953, leg. A. S. Yadav. (Fig : 13, 14, 15 and 16)

This rust was first collected by Hobson in Belgaum, Bombay on *Flueggea virosa*. The host was misidentified as *Capparis* sp., but Mundkur and Thirumalachar (Mycol. pap. No. 16, C. M. I., p. 11-12, 1946) pointed out that the host was *Flueggea virosa*. Only telial stages were described for the rust in India, but the pycnial, aecial and uredial stages have been reported from the Philippines. The rust has been collected only once in India by Hobson which constituted the type for the genus *Masseeella*. While no material exists in Indian herbaria, three leaves constituting the type are deposited at the Royal Botanic Gardens, Kew. From the present study it appears, that *M. capparidis* is of common occurrence in the mid-hills of Bihar.

12. *Ravenelia evernia* Syd. Ann. Mycol. 31 : 87, 1933.

Hab. On leaves of *Mimosa himalayensis*, Rajgir hills, 10-10-1952, leg. A. S. Yadav. The rust has previously been reported from India on *Mimosa hamata*.

13. *Coleosporium clematidis* Barclay.

Hab. on leaves of *Clematis gouriana*, Parasnath, 11-4-1954, leg. A. S. Yadav. The rust has previously been recorded from the same locality on *C. nutans*.

14. *Puccinia phyllocladiae* Cke.

On cladodes of *Asparagus*, sp., Netarhat, 30-12-1952; leg. M. J. Thirumalachar. (Figs. 17 and 18).

15. *Phakopsora parasnathii* Yadav & Thirumal.

Uredia unknown. Telia mostly hypophyllous, minute, dark-brown, scattered or rarely aggregate, subepidermal, appearing as indehiscent lenticular crusts, 340-580μ. long, 80-204μ. high ; teliospores oblong or irregularly cuboid, golden-brown, 10-16$\times$16-24μ.

Hab. on leave of dicot host (Guttiferae?), Parasnath, 11-4-1954, leg. A, S. Yadav. (Fig. 19).

Uredia ignota. Telia plerumque hypophylla, minuta, fusco-brunnea, sparsa vel raro aggregata, subepidermalia, unita lenticularium crustarum unstar, indehiscentia, 340-580μ. longa, 80-204μ. alta ; teliosporae oblongae vel irregulariter cuboideae, aureo-brunneae, 10-16$\times$16-20μ.

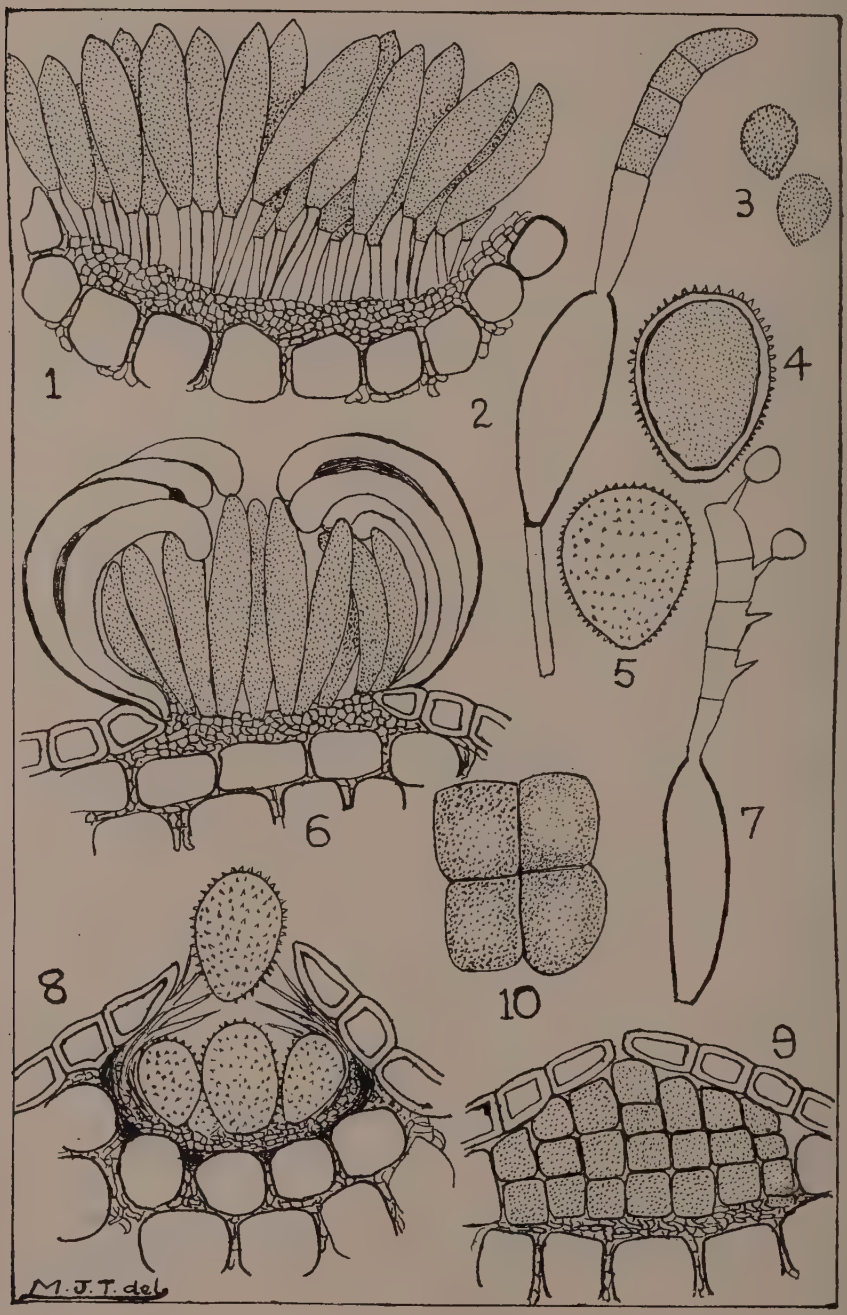
The indehiscent lenticular crusts with the teliospores developing in irregular succession from the base of the sorus also occurs in *Bubakia*. Since the uredial stages have not been observed inspite of detailed examination, the allocation of the species to *Phakopsora* is tentative.

In conclusion, the senior author wishes to express his grateful thanks to Shri J. G. Srivastava for identification of some of the hosts and to Dr, R. P. Roy for kind encouragement.

Science College, Patna
and
Poona, Bombay.

Explanation to Figs.

Fig. 1. Telium of *Maravalia milletiae* × 750
Fig. 2. Germinating teliospore × 1000
Fig. 3. Sporidia × 1500
Figs. 4-5. Urediospores × 1250
Fig. 6. Telium of *Olivea colebrookiana* × 750
Fig. 7. Germinating teliospore × 1000
Fig. 8. Uredium of *Cerotelium bauhiniae* × 750
Fig. 9. Telium × 500
Fig. 10. Groups of teliospores × 1250
Fig. 11. Telium of *Cerotelium kirganeliae* × 500
Fig. 12. Germinating teliospore × 1250
Fig. 13. Leaf of *Flueggea virosa* with spore tendrils of *M. capparidis* × 1
Fig. 14. Urediospore of *M. Capparidis* × 1000
Fig. 15. Portion of spore tendril × 400
Fig. 16. Teliospore × 1000
Figs. 17-18. Teliospore of *Puccinia phyllocladiae* × 750
Fig. 19. Section throug telium of *Phakopsora parasnathii* × 750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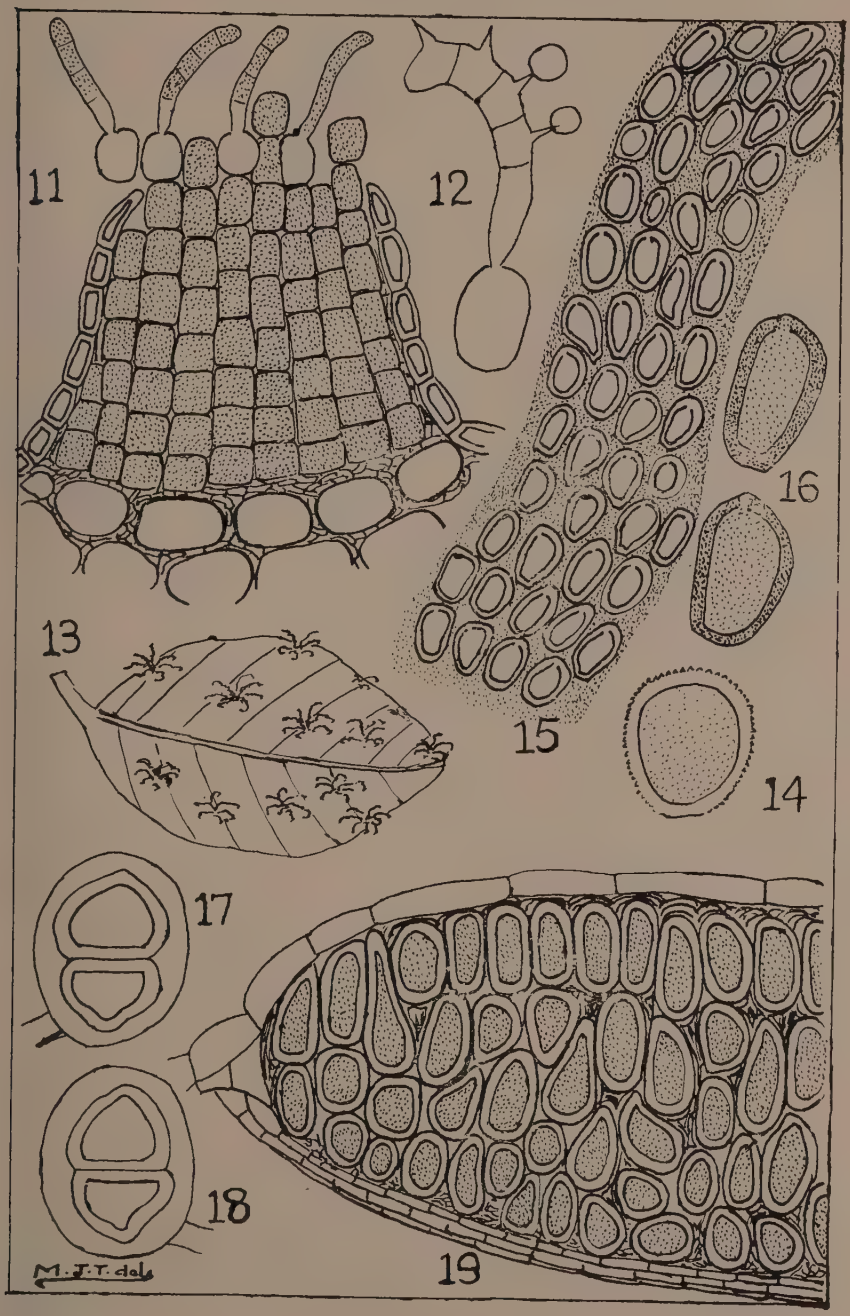
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THE MYXOMYCETES OF THE MUSSOORIE HILLS—I

K. S. THIND AND H. S. SOHI

(Accepted for publication, October 12, 1955)

The Panjab University Botany Department arranges a Botanical Excursion each year to the Mussoorie Hills, under the leadership of Prof. P. N. Mehra, to make a comprehensive study of the Cryptogamic Flora of that region. The taxonomic studies of the Myxomycetes is a part of the Fungal Flora under that programme.

Lodhi (1934) published, "Indian Slime-Moulds (Myxomycetes)", from the Panjab University Botany Department, Lahore. In all 54 species of Myxomycetes are reported in this publication and this list includes those described by Lister (1924) and Bruhl and Gupta (1927). Recently Agnihothrudu, 1954, 1954b, and 1955 has reported 12 species of slime moulds from Southern India. Out of these 7 appear to be new records for India. After a lapse of about two decades the taxonomic studies of Indian slime moulds are again resumed from this Department on a more extensive basis. These studies are confined to the Mussoorie Hills which have already yielded a rich collection of slime moulds.

This paper deals with the taxonomy of 7 species belonging to the family Physaraceae of the order Physarales. Four species—*Physarum bivalve* Pers., *P. compressum* Alb. & Schw., *P. citrinum* Schum., *P. nucleatum* Rex are new records in India while strongly plasmodiocarpous form of *Physarum cinereum* (Batsch) Pers. and sessible form of *Craterium leucocephalum* (Pers.) Ditm. are observed for the first time in India. The classification as proposed by Martin, 1949, in North American Flora 1 : 1 (Myxomycetes) has been followed in the present study.

The writers are deeply indebted to Dr. G. W. Martin of the State University of lowa, U. S. A. for help in the identification of the species and Prof. P. N. Mehra for valuable criticism and encouragement. They are also thankful to Mr. Balram Khanna for making illustrations of fructifications.

1. *Physarum cinereum* (Batsch) Pers.

Fructifications mostly sporangiate, sometimes forming small plasmodiocarps. *Sporangia* gregarious or scattered, sessile, ash coloured to whitish gray, globose or elongated, up to 0.45 mm. in diameter. *Hypothallus* absent. *Peridium* single, thin, membranous, grayish, covered over with calcarious matter which consists of rounded crystals of lime. *Dehiscence* irregular, upper portion of the peridium usually ruptures by irregular longitudinal slits so that the lower persistant portion of the peridium possesses a lobed appearance. *Columella* none. *Capillitium* consists of dense network of nodes and

internodes. Nodes spherical to irregular in shape and size, white, calcarious. Nodes interconnected by short, slender, hyaline, non-calcarious internodes. *Spores* black in a mass, violaceous under the miscroscope, globose, very minutely verrucose, 8.0-9.8μ in diameter. *Plasmodium* greenish yellow, forming an irregular network over the substratum. (Pl. I, fig. 1-2; Text fig. 1).

Collected on dead leaves of *Quercus incana* Roxb., living leaves of *Strobilanthes* sp. and living mosses, Tehri Road, Mussoorie, Aug., 1952, 1. On dead leaves of *Q. incana*, Dhobi Ghat, Mussoorie, Aug., 1952, 2. The dead leaves as well as living leaves and living mosses are not hosts but merely substrata upon which the plasmodium has crawled.

Fructifications of collection 2 are mostly plasmodiocarpous in contrast to the mostly sporangiate ones of 1 and its spores are slightly larger than those of 1. Otherwise, both the collections resemble closely. Collection 2 is regarded here as strongly plasmodiocarpous form of *P. cinereum* with spores mostly from 9 to 10 μ in diameter.

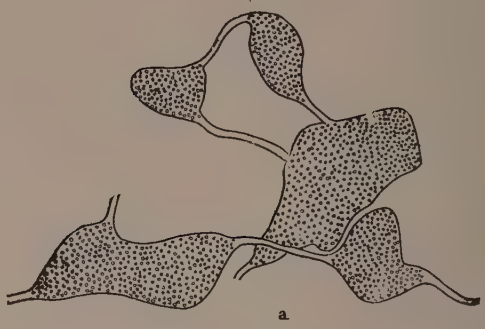
2. *Physarum bivalve* Pers.

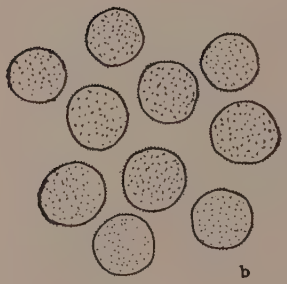
Fructifications plasmodiocarpous. *Plasmodiocarps* long or short, sinuous or in short curved segments, thick, laterally compressed, *i.e.*, raised like plates, sometimes reduced to sporangial forms, dull yellowish, up to 0.5 mm. in diameter. *Hypothallus* absent. *Peridium* plainly double. Outer peridium dull yellowish on the outside and white from the inside, thick, calcarious; inner peridium thin and grayish. *Dehiscence* regular, by a longitudinal slit in the summit of fructifications. The outer peridium after splitting longitudinally looks like two halves of a water mussel (a bivalve mollusc), the inner peridium remains intact longer and later on ruptures irregularly. *Columella* absent. *Capillitium* profusely developed, a dense mass of white calcarious nodes which are spherical or irregular, internodes are developed poorly and the nodes may be interconnected by other nodes or by internodes. *Spores* black in a mass, dark violet under the miscroscope, globose, somewhat angular, and thick walled, all of which may be due to unduly rapid drying in maturation, minutely and profusely verrucose, with a clear smoother area, 9-11 μ in diameter. (Pl. I, fig. 3; Text fig. 2).

Collected on dead leaves of *Quercus incana* and other plants, Dhobi Ghat, Mussoorie, Sept., 1952, 3. New record in India.

3. *Physarum compressum* Alb. & Schw.

Fructifications sporangiate. *Sporangia* gregarious or scattered, stipitate, laterally compressed, variable in shape, obovate, reniform, or fan shaped, entire or lobed, ashen gray, or ashen white, 1—1.35×0.83 mm. Stipe long, erect, slightly tapering above, irregularly furrowed, grayish white, calcarious, up to 0.67 mm. in height. *Hypothallus* dark brown, rotate. *Peridium* single, ashen gray, prominently calcarious, calcarious matter white, mealy or in the form of minute whitish scales. *Dehiscence* irregular, peridium rupturing any-





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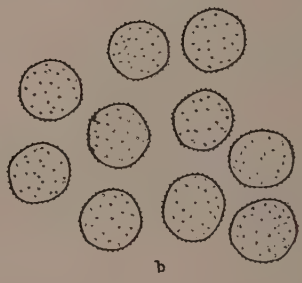

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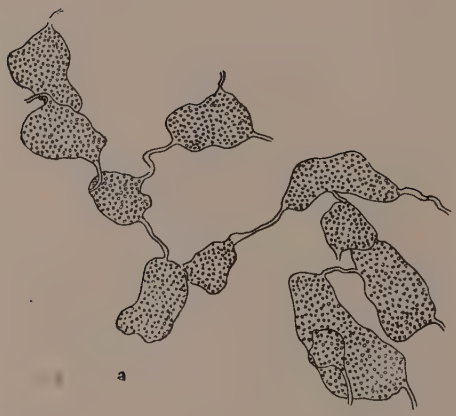

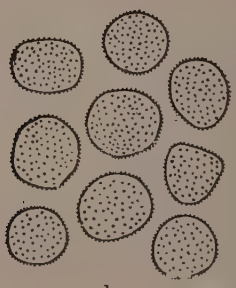

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where to expose and release the spores. *Columella* absent. *Capillitium* abundant, a dense network of nodes and internodes. Nodes large, irregular in shape and size, whitish, calcarious, calcarious matter consisting of rounded crystals of lime. Nodes interconnected with one another by long slender, hyaline, thread-like, noncalcarious internodes. *Spores* black in a mass, dark violet under the microscope, thick-walled, mostly oval to ovoid, sometimes triangular with rounded edges, or globose to subglobose or ellipsoidal, distinctly and profusely verrucose, 9.5—13 μ in diameter. The variable shape of the spores seems to be due to rapid drying in maturation. (Pl. I, fig. 4; Text fig. 3).

Collected on dead bark of *Rhododendreon arboreum* Smith and living mosses, Company Garden, Mussoorie, Sept., 1952, 4. New record in India.

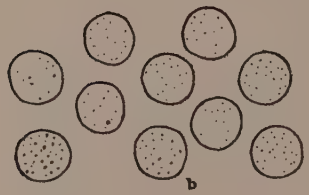
4. *Physarum melleum* (Berk. & Br.) Massee

Fructifications sporangiate. *Sporangia* gregarious, stipitate, mostly orange or orange red, sometimes dull yellow, globose to subglobose, marked by irregular ridges, 0.5—0.52×0.46 mm. Rarely the bodies of the neighbouring sporangia or their stipes may be connate. Stipe erect, white, clacarious, thick below, tapering above, grooved throughout, up to 0.4 mm. long. *Hypothallus* well developed. white, calcarious, rotate. *Peridium* thin, orange red, marked by rounded crystals of lime. *Dehiscence* irregular, by rupturing of the peridium anywhere. *Columella* small but conspicuous, just protruding into the sporangial cavity, white *i.e.*, concolorous with the stipe. *Capillitium* a network of nodes and internodes. Nodes few, yellow, spherical to irregular in shape and size, calcarious. Nodes interconnected with one another by hyaline, slender, threadlike flexuous, noncalcarious internodes. *Spores* black in a mass, pale violet under the microscope, globose to subglobose, inconspicuously verrucose, 6.5—9.5 μ in diameter. *Plasmodium* orange and like an irregular network. (Pl. II, fig. 5 ; Text fig. 4)

Collected on living mosses and on dead leaves of *Quercus incana* and other plants, Mussoorie, Aug., 10, 1952, 5. On dead leaves of *Q. incana* and other plants and on living leaves of *Gerbera* sp., Nala Pani, Dehradun, Aug. 7, 1954, 6.

5. *Physarum citrinum* Schum.

Fructifications sporangiate. *Sporangia* gregarious, scattered, stipitate, sulphur yellow with a greenish tinge, globose or subglobose, 0.4—0.5 mm. in diameter. Stipe long, erect, thick and bulbous below, very much narrowed down above, lower thick and bulbous portion white, while the upper thin portion is light yellow. Stipe up to 0.57 mm. in height, *i.e.*, nearly equal to the size of the sporangium. *Hypothallus* none. *Peridium* thin, sulphur yellow, calcarious, covered by the rounded crystals of lime. *Dehiscence* irregular, by rupturing of the peridium at any place. *Columella* small, conical, yellow. *Capillitium* a network of nodes and internodes. Nodes sulphur yellow with



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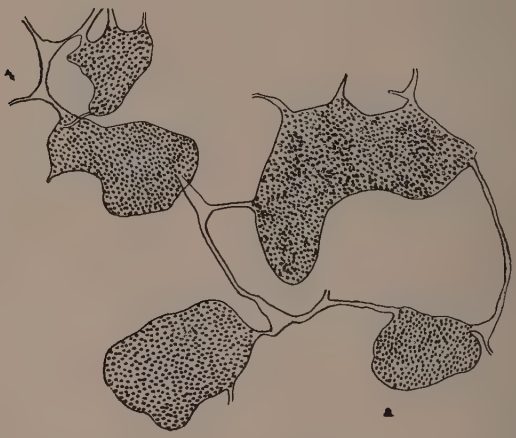

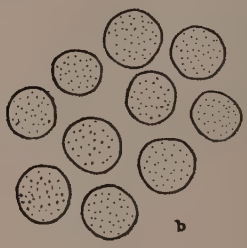

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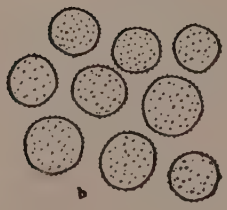

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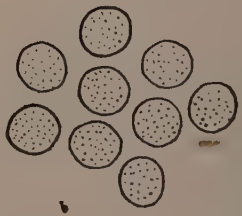

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a greenish tinge, spherical to irregular in shape, calcarious. Nodes interconnected with one another by slender, hyaline, noncalcarious internodes. *Spores* black in a mass, violaceous under the microscope, globose to subglobose, minutely verrucose, 8—8.5 μ in diameter. (Pl. II, fig. 6; Text fig. 5).

Collected on dead leaves of *Quercus incana* and other plants, Burning Ghat, Mussoorie, Sept. 10, 1951, 7. New record in India.

6. *Physarum nucleatum* Rex.

Fructifications sporangiate. *Sporangia* gregarious or densely crowded, stipitate, grayish white, globose, upright or nodding, 0.37—0.40 mm. in diameter. Stipe long, erect or bending, thick below, tapering above, dark brown below and yellowish brown above, grooved throughout, up to 2.50 mm. in height, i. e., about six times the height of the sporangium. *Hypothallus* yellowish, rotate. *Peridium* thin, black, calcarious looking grayish white due to lime deposit. *Dehiscence* by rupturing of the upper portion of the peridium, lower portion remaining persistant. *Columella* none. *Pseudo-columella* present. *Capillitium* a dense, delicate network of nodes and internodes. Nodes spherical to irregular in shape and size, white, calcarious, calcarious matter consisting of rounded crystals of lime. Nodes interconnected with one another by long, slender, hyaline, threadlike, noncalcarious internodes. Each sporangium has in its centre a prominent, white, spherical mass of lime 'nucleus' which is not extended below to the stipe. *Spores* black in a mass, violaceous under the microscope, rounded, minutely verrucose, 8—10μ in diameter. (Pl. II, fig. 7 ; Text fig. 6).

Collected on rotting wooden stumps and dead leaves of *Quercus incana* and other plants, Kempty Fall, Mussoorie, Aug. 29, 1952, 8. New record in India.

7. *Craterium Leucocephalum* (Pers.) Ditm.

Fructifications sporangiate. *Sporangia* mostly gregarious, may also be scattered, stipitate, whitish above, brown with a reddish tinge in the lower portion, cyathiform or turbinate, typically bell-shaped after lid is detached, ridged on the outside, ridges being prominent below. Sporangia measure 0.90—0.98×0.30 mm. Stipe long, erect, deep brown with a reddish tinge, grooved, uniform in thickness except the base where it is slightly broader, from 0.25—0.27 mm. in height. *Hypothallus* well developed, deep brown with a reddish tinge, rotate. *Peridium* single, persistent except the lid. *Dehiscence* regular, circumscissile, occurs by a distinct circular lid which covers entirely the top of the sporangium and lies usually below the slightly thickened and everted margin of the cup, later on the lid falls off leaving the bell shaped sporangium below. Lid is whitish on the outer surface due to the calcarious deposit. *Columella* none. *Pseudocolumella* strongly developed like a columella along the axis of the cup, snow white, calcarious, and formed by the larger nodules (or nodes) massed together along the middle of the fructifications. *Capillitium* a dense network of nodes and internodes. Nodes large, conspicuous, persistent, spherical to irregular in shape and size, white, calcarious, calcarious matter

consisting of rounded crystals of lime. Nodes interconnected by slender, hyaline, thread-like, noncalcarious internodes. *Spores* black in a mass, violaceous under the microscope, globose, minutely and inconspicuously verrucose, 7—8μ in diameter. (PI. II, fig. 8-9 ; Text fig. 7).

Collected on dead leaves and branches of *Quercus incana*, *Saccharum* species and other plants, Kempty Fall, Mussoorie, Aug., 1952, 9.0n dead leaves and branches of *Q. incana*, Lal Tibba, Mussoorie, Aug. 24, 1952, 10.

The fructifications of collection 10 are slightly darker, sessile, and reticulately or irregularly ridged at the top (upper side of the lid) which is smooth in 9. Otherwise, both the collections resemble closely. Collection 10 is regarded here as a sessile form of *C. leucocephalum*, but spores are more finely warted, as is usually in this species. *Plasmodium* was observed only in collection 10 and it is yellow and forms a network over the substratum.

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PLATE I

PLATE II

Explanation of Plates

Plate I

Fig. 1. Mostly sporangiate fructifications of *Physarum cinereum*, X20.
Fig. 2. Strongly plasmodiocarpous form of *P. cinereum*, X20.
Fig. 3. Laterally compressed plasmodiocarps of *P. bivalve*, X20.
Fig. 4. Laterally compressed sporangia of *P. compressum*, X20.

Plate II

Fig. 5. Sporangia of *P. melleum*, X20.
Fig. 6. Sporangia of *P. citrinum*, X20.
Fig. 7. Sporangia of *P. nucleatum*, X20.
Fig. 8. Sporangia of *Craterium leucocephalum*, X20.
Fig. 9. Sessile form of *C. leucocephalum*, X20.

Explanation of Text-Figures

Fig. 1. *Physarum cinereum.* (a) Capillitium, X380., (b) Spores, X880.
Fig. 2. *P. bivalve.* (a) Capillitium, X380., (b) Spores, X880.
Fig. 3. *P. compressum.* (a) Capillitium, X380., (b) Spores, X880.
Fig. 4. *P. melleum.* (a) Capillitium, X380., (b) Spores, X880.
Fig. 5. *P. citrinum.* (a) Capillitium, X380., (b) Spores, X880.
Fig. 6. *P. nucleatum.* (a) Capillitium with minute and less numerous nodes, X880., (b) Spores, X880.
Fig. 7. *Craterium leucocephalum.* (a) Capillitium, X380., Spores, X880.

TWO NEW RECORDS OF PHYTOPATHOGENIC BACTERIA FROM BOMBAY STATE

V. V. Bhatt and M. K. Patel

(Accepted for publication, October 14, 1955)

In the present paper, an account of two new species of *Xanthomonas* previously described by the authors* in a preliminary note is given in detail.

(1) *Alysicarpus rugosus* DC. is a well known green fodder in Bombay State. A leaf-spot and severe blight of bacterial origin on this plant was observed at Jalgaon during the rainy season of 1953.

The pathogen was easily isolated by the usual poured plate method and a single colony, subcultured and maintained on neutral potato dextrose agar, proved pathogenic when inoculated on healthy plants of *A. rugosus.*

Symptoms

The pathogen on *A. rugosus* produces a few, small, round, water-soaked spots, measuring about 0.5-1.0 mm. mostly at the periphery of the leaf. On further development of the disease, the spots with pale yellow parched centre and brown periphery produce blight in the leaves (Fig. 1.) which turn yellow, curve and shed. Although the blight symptoms progress along the veins, the pathogen does not become vascular. From the symptomatical point of view, the pathogen resembles *Xanthomonas vignicola* Burkholder which produces similar symptoms in *Vigna catjang* Walp. Besides infecting leaves, it produces canker on the injured tender stem.

Morphology and Cultural Characters

A 48-hour old culture of the pathogen when stained was found to be short single rods, $0.6 \times 2.0\ \mu$ in size. Gram negative, capsulated, non-spore former and motile by single polar flagellum (Fig. 2). Agar colonies were circular with entire margin, smooth, raised, butyrous and pale lemon yellow (R) on potato dextrose and empire yellow (R) on nutrient dextrose. It made good growth on yeast-dextrose-chalk and Krumweide's agars but failed to grow in synthetic nitrate and Czapek's media. Optimum temperature for growth was 27°—30°C. and thermal death point about 51°C.

Biochemical Characters

The culture liquefied gelatin; hydrolysed starch; digested casein; peptonised litmus milk and reduced it with slight acid reac-

Curr. Sci. **23** : 165, 1954.

tion ; completely liquefied Loeffler's blood serum in 8 days ; produced hydrogen sulphide and ammonia from peptone ; did not produce indol from tryptophane, and nitrite and ammonia from nitrate ; tolerated sodium chloride upto 3 per cent and pH from 4 to 9 ; made no growth in Cohn's ammonium tartarate and Clara's asparagine media but grew well in Uschinsky's glycerol ammonium lactate medium with alkaline reaction and Fermi and Montesano's glycerol ammonium tartarate medium with acid reaction ; produced good growth and acid without gas from arabinose, galactose, levulose, rhamnose, xylose, dextrose, lactose, maltose, sucrose, starch, glycerol, mannitol, raffinose and alkali from acetic, citric aud lactic acids but no growth in salicin, cellulose and acids like benzoic, formic, oxalic, salycilic, tannic and tartaric ; utilised nitrogen from inorganic ammoniacal compounds but not from nitrates of potassium, magnesium and sodium ; utilised nitrogenous organic compounds like alanine, asparagine, aspartic acid, creatin, creatinine, cystine, guanidine hydrochloride, glycine, glucocyamine, leucine, methionine, norvaline, ornithine hydrobromide, phenyl alamine, serine, threonine, tryptophane, tyrosine, urea and valine in presence of dextrose while glutamic acid and proteose peptone supplied both the carbon and nitrogen for growth.

Host Range

The pathogen was restricted to *A. rugosus* only. It did not infect *Acacia arabica* Willd., *A. catechu* Willd., *A. decurrens* Willd., *A. melanoxylon* Br., *Alysicarpus bupleurifolius* DC., *A. hamosus* Edgew., *A. longifolius* W. & A., *A. monilifer* DC., *A. pubescens* Law., *A. tetragonolobus* Edgew., *A. vaginalis* DC., *Arachis hypogaea* L., *Butea frondosa* Konig., *Caesalpinia pulcherrima* Swartz., *C. sepiaria* Roxb., *Cassia alata* L., *C. didymobotrya* Fraesen, *C. hirsuta* L., *C. siamea* Lam., *Centrosema pubescens* L., *Cicer arietinum* L., *Crotolaria anagyroides* H. B. & K., *C. juncea* L., *C. striata* DC., *Dolichos biflorus* L., *D. lablab* L., *Indigofera argenta* L., *I. arrecta* Benth., *I. glandulosa* Willd., *I. tinctoria L.*, *Lathyrus odoratus* L., *L. sativus* L., *Leucaena glauca* L., *Melilotus indica* All, *Moringa pterygosperma* Gaertn., *Phaseolus aconitifolius* Jacq., *P. angularis* Wight., *P. coccineus* Lam., *P. lunatus* L., *P. mungo* var. *radiatus* L., *P. vulgaris* L., *Pisum arvense* L., *Poinciana regia* Bojer, *Pueraria phaseoloides* Borth., *Sesbania aculeata* Poir., *Tephrosia candida* DC., *T. purpurea* Pers. *Trifolium alexandricum* L., *Trigonella foenum-graecum* L., *Vigna sinensis* Endl. and the other hosts given elsewhere in this paper.

Since no bacterial disease has so far been reported on this host and as the pathogen is restricted to *A. rugosus* only, it is proposed to assign the bacterium a new name, *Xanthomonas alysicarpi.*

Xanthomonas alysicarpi Bhatt and Patel sp. nov. On leaves of *Alysicarpus rugosus* DC. Found at Jalgaon in monsoon, 1953. Leg. M. K. Patel.

Short rods ; single polar flagellum ; Gram negative ; capsulated ; agar colonies smooth, circular, raised. butyrous and yellow ; gelatin liquefied ; starch hydrolysed ; casein digested ; milk peptonised and

litmus reduced; hydrogen sulphide and ammonia produced from peptone; nitrite not produced from nitrate; no growth in synthetic nitrate and Czapek's media; acid without gas from arabinose, dextrose, lactose, sucrose and starch; no growth in salicin; optimum temperature for growth 27°—30°C.; thermal death point about 51°C.

* * * * *

(2) *Bridelia hamiltoniana* Wall., a common shrub growing on waste lands along the Western Coast of India, was found parasitised by bacteria at Ambarnath (Thana) in July, 1953. The bark of the plant is used as an astringent while the leaves are used as fodder.

Small, shining bacterial colonies appeared after 48 hours incubation at 30°C. when isolated by usual poured plate method and proved pathogenic.

Symptoms

The pathogen on *B. hamiltoniana* produces a few to numerous minute, water-soaked spots clearly visible on the lower surface, measuring initially 0.3 -0.5 mm., fairly well distributed all over the rhomboid leaf. High atmospheric humidity and continuous rains are congenial for the development of the disease. With the progress of the disease, the spots with small yellow halo around increase in size to 1.0—1.5 mm., become angular, dark brown to jet black and when coalescent produce leaf curl and parching of the leaf tissues which may shed off leaving shot holes (Fig. 3). Bacterial ooze in the form of small shining beads is found on the lower surface of the leaf. The pathogen infects leaves only.

Morphology and Cultural Characters

A young culture of 48 hours when stained was found to be short rods, mostly single, 0.5×1.7 μ in size, motile by single polar flagellum. Gram negative, capsulated and non-spore former. It produced circular, entire, smooth, raised, butyrous, barium yellow (R) colonies on potato dextrose and pinard yellow (R) colonies on nutrient dextrose agars. It made good growth on yeast-dextrose-chalk and Krumweide's agars but no growth in synthetic nitrate and Czapek's media. Optimum temperature for growth was 27°—30°C, and thermal death point about 51°C.

Biochemical Characters

Gelatin liquefied; starch hydrolysed and casein digested; milk peptonised and litmus reduced with slight acidity; Loeffler's blood serum completely liquefied in 8 days; hydrogen sulphide and ammonia produced from peptone; indol not produced from tryptophane; nitrite and ammonia not produced from nitrate; sodium chloride tolerated upto 3 per cent and pH from 4 to 9; Cohn's and Clara's media did not support the growth while good growth and alkalinity observed in Uschinsky's and fair growth and fair acidity in Fermi and Montesano's media; good growth and acid without gas from arabinose, galactose, levulose, rhamnose, xylose, dextrose, lactose, maltose,

sucrose, starch, mannitol ; alkali from acetic, citric and lactic acids ; slight to fair growth in dulcitol, glycerol. raffinose and no growth in salicin, cellulose and acids like benzoic, formic, oxalic, salycilic, tannic and tartaric ; nitrogen from inorganic ammoniacal compounds utilised but that from nitrates of potassium, magnesium and sodium not utilised ; organic nitrogenous compounds like alanine. asparagine, aspartic acid, creatin, creatinine, cystine, guanidine hydrochloride, glycine, glucocyamine, leucine, methionine, norvaline. ornithine hydrobromide, phenyl alamine, sarine, threonine, tryptophane, tyrosine, urea and valine utilised in presence of dextrose while glutamic acid and proteose peptone supplied both the carbon and nitrogen for growth.

Host Range

The pathogen infects only *Bridelia hamiltoniana* and not *Aegle marmelos* Corr., *Acalypha* sp., *Alysicarpus rugosus* DC., *Amaranthus viridis* L., *Begonia* spp., *Brassica oleracea* L., *Capsicum annuum* L., *Cajanus cajan* Millsp., *Cassia tora* L., *Citrus aurantifolia* Sw. and *Clerodendron phlomoides* L., Croton spp., *Cyamopsis tetragonoloba* (L.) Taub., *Desmodium diffusum* DC., *D. gangeticum* DC., *Erythrina indica* Lam., *Euphorbia geniculata* Ort., *E. hypericifolia* L., *E. pulcherrima* Willd., *E. splendense* Boj., *Gossypium herbaceum* L., *Ipomoea muricata* R. & Sch., *Lawsonia alba* Lam., *Manihot esculenta* Crantz., *Medicago sativa* L., *Piper betle* L., *Pisum sativum* L., *Ricinus communis* L., *Sesbania aegyptiaca* Prain., *Soja max* (L.), Piper, *Stizolobium deeringianum* Bort., *Tamarindus indica* L., *Tectona grandis* L., *Trichodesma zeylanicum* R. & Br., *Vigna catjang* Walp., *Xanthium strumarium* L. and the other hosts of *Xanthomonas* spacies in Bombay State.

As the pathogen under study is indistinguishable from the other members of the genus *Xanthomonas* in important taxonomic characters but being highly specific to its own host, it is proposed to assign it a specific name *Xanthomonas brideliae.*

Xanthomonas brideliae Bhatt and Patel sp. nov. On leaves of *Bridelia hamiltoniana* Wall. Found at Ambarnath (Thana), Bombay in July, 1953. Leg. V.V. Bhatt.

Short rods ; single polar flagellum ; Gram negative ; capsulated ; agar colonies smooth, circular, raised, butyrous and yellow ; gelatin liquefied ; starch hydrolysed ; casein digested ; milk peptonised and litmus reduced ; hydrogen sulphide and ammonia produced from peptone ; nitrite not produced from nitrate ; no growth in synthetic nitrate and Czapek's media ; acid without gas from arabinose, dextrose, lactose, sucrose and starch ; no growth in salicin ; optimum temperature for growth 27°—30°C. ; thermal death point about 51°C.

Summary

Two new species of phytopathogenic bacteria parasitising *Alysicarpus rugosus* and *Bridelia hamiltoniana* are reported.

Alysicarpus rugosus

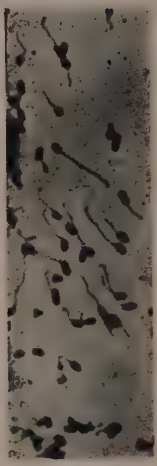

Fig. 2

X. alysicarpi

Fig. 3

Bridelia hamiltoniana

Fig. 2

X. alysicarpi

From the morphological, cultural and biochemical study, it is observed that the pathogens belong to the genus *Xanthomonas*. Out of 87 hosts tried so far, it is observed that the pathogens isolated from *A. rugosus* and *B. hamiltoniana* respectively are pathogenic to their own susceptˢ and thereby allotted a status of *novum species* to each.

Disease specimens and pathogenic cultures are deposited in the herbarium and culture collection of the Plant Pathologist to Government, Bombay State, College of Agriculture, Poona, India. The cultures are also deposited with Dr. M. P. Starr, Department of Bacteriology, University of California, Davis, California, U. S. A.

Plant Pathological Lab.,
College of Agriculture,
Poona 5.

Explanation of Figures

Fig. 1. No. 1, 2, 3, 4 and 5 showing the progressive development of blight symptoms on the leaves of *Alysicarpus rugosus*.

Fig. 2. Single polar flagellum of *Xanthomonas alysicarpi*.

Fig. 3. Angular dark brown to jet black spots with small yellow halo around on the leaves of *Bridelia hamiltoniana*.

PHYTOPATHOLOGICAL SURVEY OF THE HYDERABAD STATE

SYED VAHEEDUDDIN.

(Accepted for publication, October 12, 1955)

The Phytopathological survey of the Hyderabad State was started by the author in the year 1938-39 and was completed in 1945-46. The system of survey followed was to select at least three taluqs at random in each district and at least three villages in each taluq. In each village surveyed a minimum of six fields of different cultivated crops and as many fruit gardens as were available were surveyed. Most of the villages were visited only once during the period of survey.

Name of organism	Host	Common name of the disease	Locality
PHYCOMYCETES			
1. *Cystopus candidus* (Pers.) Lev.	*Portulaca* & Raddish	White rust	Hyderabad
2. *Pythium graminicolum* Subramaniam	Wheat	Foot rot	Nanded.
3. *Pythium aphanidermatum* (Eds.) Fitz.	Papaya & Ginger	Foot rot & Soft rot	Vikarabad.
4. *Pythium debaryanum* Hesse	Tobacco	Damping off	Mahabubnagar and Warangal.
5. *Phytophthora colocasiae* Rac.	Colocasia	Blight	Hyderabad.
6. *Phytophthora palmivora* Butler	Palms	Bud rot	Nalgonda and Hyderabad.
7. *Phytophthora parasitica* Dast.	Castor	Blight	Hyderabad.
8. *Phytophthora parasitica* Dast.	Sesamum	Leaf spot	Nizamabad.
9. *Peronospora brassicae* Gaumann	Mustard	Downy mildew	All districts
10. *Plasmopara viticola* Berk. & Curt.	Grapevine	do	do
11. *Sclerospora graminicola* (Sacc.) Sch.	Bajra	do	Raichur and Bider.
12. *Sclerospora sorghi* Weston & Uppal	Jowar	do	Raichur.
13. *Sclerospora maydis* (Rac.) Butler	Maize	do	Karimnagar.
14. *Rhizopus artocarpi* Rac.	Jack fruit	Foot rot & Soft rot	Hyderabad
ASCOMYCETES			
1. *Balansia oryzae* (Syd.) Nars. et Thirum.	Paddy	Udbatta disease	Medak and Adilabad.
2. *Cochliobolus miyabeanus* (Ito & Kurib.) Drechsler ex Dastur	do	Leaf blight	Hyderabad.
3. *Claviceps purpurea* (Fr.) Tul.	Rye	Ergot	Warangal
4. *Elsinoe ampelina* Shear	Grapevine	Anthracnose	All Dists.

	Name of organism	Host	Common name of the disease.	Locality
5.	*Erysiphe cichoracearum* DC.	Cucurbits, Mango, Bhendi & Tobacco.	Powdery mildew.	Raichur, Mahabubnagar & Nizamabad.
6.	*Erysiphe polygoni* DC.	Peas. Pulses Coriander.	do	do
7.	*Erysiphe* sp.	Mentha (Mint)	do	Hyderabad
8.	*Glomerella cingulata* (Stonem.) Spauld. and von Schrenk.	Betel leaf	Anthracnose	Latur.
9.	*Glomerella gossypi* (Southw.) Edgerton	Cotton	do	Nanded & Hyderabad.
10.	*Leveillula taurica* (Lev.) Arn.	Garlic & Cluster Bean	Powdery mildew	Gulberga.
11.	*Mycosphaerella arachidicola* W. A. Jenkins	Groundnut	Tikka	All districts
12.	*Mycosphaerella berkeleyii* W. A. Jankins	do	do	do
13.	*Physalospora tucumanensis* Speg	Sugarcane	Red rot	Warangal & Nizamabad.
14.	*Phyllachora cynodontis* (Sacc.) Niessl.	Dub	Leaf spot	Warangal.
15.	*Rosellinia necatrix* (Hartig) Berl.	Grapevine	Root rot	Hyderabad.
16.	*Sphaerotheca pannosa* (Wallr.) Lev.	Rose	Powdery mildew	Aurangabad.
17.	*Taphrina maculans* Butler	Turmeric	Leaf spot	Bidar & Hyderabad.
18.	*Uncinula necator* (Schw.) Burr.	Grapevine	Powdery mildew	Hyderabad & All Dists.
19.	*Uncinula tectoni* Salmon	Teak	do	Hyderabad & Nizamabad.
	BASIDIOMYCETES			
1.	*Sphacelotheca cruenta* (Kuehn) Potter	Jowar	Loose smut	Aurangabad.
2.	*Sphacelotheca sorghi* (Link) Clinton	do	Grain smut	All Dists.

3.	*Sphacelotheca reiliana* (Kuehn) Clinton	do	Head smut	Aurangabad & Gulberga.
4.	*Tolyposporium ehrenbergii* (Kuehn) Pat.	do	Long smut	Hyderabad & Nizamabad.
5.	*Tolyposporium penicillariae* Bref	Bajra	Smut	Raichur.
6.	*Ustilago crameri* Koernicke	Setaria	do	do
7.	*Ustilago cynodontis* P. Hennings	Dub	Smut	Hyderabad and Nizamabad.
8.	*Ustilago nuda* (Jensen) Rostr.	Barley	Loose smut	Zaheerabad.
9.	*Ustilago scitaminea* Syd.	Sugarcane	Whip smut	All Districts.
10.	*Ustilago shiraiana* P. Henn.	Bambusa	Smut	Adilabad.
11.	*Ustilago tritici* (Pers.) Rostr.	Wheat	Loose smut	Parbhani.
12.	*Ustilago panici miliacei* (Pers.) Wint.	Italian Millet	Smut	Hyderabad.
13.	*Melampsora lini* (Pers.) Lev.	Linseed	Rust	Hyderabad.
14.	*Melampsora ricini* (Biv.) Pass.	Castor	do	Hyderabad and Zaheerabad.
15.	*Cerotelium fici* (Cast.) Arth.	Fig	Rust	All Districts.
16.	*Cerotelium desmium* (Berk. & Br.) Arth.	Cotton	do	Parbhani.
17.	*Puccinia graminis tritici* (Pers.) Eriks. & Henn.	Wheat and Barley.	Stem rust	All Districts.
18.	*Puccinia helianthi* Schwein	Sun flower	Rust	Hyderabad.
19.	*Puccinia kuehnii* (Krueg.) Butler	Sugarcane	do	Bodhan.
20.	*Puccinia purpurea* Cooke.	Jowar	Rust	Gulberga.
21.	*Puccinia maydis* Bereng.	Maize	do	Karimnagar.
22.	*Puccinia penniseti* Zimmerm.	Bajra	do	Aurangabad.
23.	*Olivea tectonae* (Raciborski) Thirumalachar	Teak	do	Nizamabad and Hyderabad.
24.	*Angiopsora ampelopsidis* (Dietal & Sydow) Thirum.	Grapevine	do	Nizamabad.
	FUNGI IMPERFECTI			
1.	*Alternaria solani* (Ell. & Mart.) Jones and Grout.	Potato	Early blight	Hyderabad Nizamabad and Bider.

	Name of organism	Host	Common name of the disease	Locality
2.	*Alternaria palandui* Ayyangar.	Garlic	Leaf spot	Parbhani.
3.	*Cephalosporium sacchari* Butler.	Sugarcane	Wilt	Bider & Nizamabad.
4.	*Cercospora cucurbitae* Ell. & Ev.	Cucurbits	Leaf spot	Gulberga.
5.	*Cercospora carthami* Sundar, & Ramkrishnan	Safflower	do	Hyderabad.
6.	*Cercospora ricinella* Sacc. & Berl.	Castor	do	Nalgonda and Mahaboobnagar.
7.	*Curvularia geniculata* (Tracy & Earle) Boedijn.	Paddy	Discolouring of grains	Hyderabad.
8.	*Curvularia pallescens* Boedijn	do	do	do
9.	*Curvularia penniseti* (Mitra) Boedijn	Bajra	Discolouring of grains	Hyderabad.
10.	*Curvularia specifera* (Bainier) Boedijn	Paddy	do	Nagar Karnool.
11.	*Fusarium bulbigenum* var. lycopersici (Brushi) Wr. & Reink.	Tomato	Wilt	Hyderabad.
12.	*Fusarium coeruleum* (Lib.) Sacc.	Potato, Tomato, Chillies & Brinjal.	do	Adilabad, Bodhan & Parbhani.
13.	*Fusarium lini* Bolley	Linseed	do	Nizamabad & Bider.
14.	*Fusarium orthoceras* App. et Wr. var. *ciceri* Padwick	Gram	do	Warangal.
15.	*Fusarium udum* Butler	Tur	do	Nanded, Bhir, Sangareddy and Mahabubnagar.
16.	*Fusarium vasinfectum* Atk.	Cotton	do	Nanded, Raichur and Parbhani.
17.	*Helminthosporium turcicum* Pass.	Maize	Leaf blight	Karimnagar.
18.	*Piricularia oryzae* Cav.	Paddy	Blast	Hyderabad, Nizamabad, Warangal and Mahabubnagar.

19. *Ramularia areola* Atkinson	Cotton	Areolate mildew	Nanded and Raichur.
20. *Sphacelia sorghi* Clinton	Jowar	Sugary disease	Gulberga.
21. *Phleospora mori* (Lev.) Sacc.	White mulberry	Leaf spot	Hyderabad.
22. *Phyllosticta mortoni* Fair.	Mango	Leaf blight	Hyderabad & Aurangabad.
23. *Phyllosticta bauhiniae* Cke.	Bauhinia	Leaf spot	Hyderabad.
24. *Pestalotia mangiferae* P. Henn.	Mango	Grey blight	do
25. *Diplodia arachidis* Petch	Groundnut	Rot	do
26. *Diplodia* sp.	Papaya	Stem rot	do
27. *Colletotrichum curcumae* (Sydow) Butler and Bisby	Turmeric	Leaf blotch	Nizamabad.
28. *Ascochyta sorghi* Sacc.	Jowar	Leaf blotch	Raichur.
29. *Macrophoma* sp.	Mango	Leaf spot	Hyderabad and Aurangabad.
30. *Graphiola phoenicis* Poiteau	Date palm	False smut	Hyderabad.
31. *Rhizoctonia solani* Kuhn.	Cotton	Rot	Nanded.
32. *Rhizoctonia* sp.	Groundnut	Root rot	Hyderabad.
33. *Sclerotium oryzae* Catt.	Paddy	Rot of paddy	do

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A BACTERIAL LEAF-SPOT DISEASE OF LOCHNERA PUSILLA*

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(Accepted for publication, October 20, 1955)

Lochnera pusilla Schum., a common weed in the cultivated fields was found to be heavily infected with a bacterial disease in the neighbourhood of Pimpri and Agricultural College Farm, Poona in June, 1954.

On isolation by usual poured plate method, small, shining, deep yellow pathogenic culture appeared after 48 hours at 30°C. which when studied for morphological, cultural, biochemical and host responses was found to be a new species of *Xanthomonas*. Artificial inoculation of healthy plants by spraying the water suspension of pure culture of the bacterium gave disease symptoms within 8 days.

Symptoms

The pathogen produces a few, round, water-soaked areas which when held against light show a small, deep brown coloured centre and pale yellow halo around, measuring 0.5—0.7 mm. With the progress of the disease, three types of symptoms are observed. In the first instance, a ring formation is seen when water-soaked areas with brown centre and pale yellow halo form a deep brown periphery around each spot measuring 1–2 mm. Secondly, sometimes the brown centre of the water-soaked areas goes on increasing so rapidly that the yellow halo around such spots is not able to keep pace with the increase in the size of the deep brown centres and ultimately, only deep brown spots measuring 1-2 mm. are seen. Lastly, in the later stage of the disease as a result of heavy infection, leaves become yellow and detach from the plants even at slight touch or breeze, the spots increase in size to about 3-4 mm., leaving a parchy centre and deep brown periphery around them (Fig. 1). Besides leaves, the pathogen infects cotyledons and pods.

Morphology and Cultural Characters

The pathogenic culture of 48 hour growth, when stained, was short single rod, 0.5—1.6μ in size, motile by single polar flagellum, Gram negative, capsulated and non-spore former. Agar colonies were circular, entire, smooth, shining, raised, butyrous, barium yellow (R) on potato dextrose and amber yellow (R) on nutrient dextrose. It also made good growth on yeast-dextrose-chalk agar, Krumweide's, synthetic nitrate and Czapek's media. Optimum temperature for growth was 27°-30°C. and thermal death point about 51°C.

*The detailed account of a short note published by the authors in Current Science, **24** : 20-21, 1955.

Biochemical Reactions

The culture liquefied Frazier's as well as plain gelatin ; equally hydrolysed starches like corn, potato, pulse, rice and wheat ; digested casein ; peptonised milk and reduced litmus with acid reaction ; slightly liquefied Loeffler's blood serum in 15 days; produced hydrogen sulphide and ammonia from peptone, indol from tryptophane, and nitrite and ammonia from nitrate ; tolerated sodium chloride upto 3 per cent and pH from 4.3 to 9.5 ; made no growth in Cohn's ammonium tartarate and Clara's asparagine media but grew well with alkaline reaction in Uschinsky's glycerol ammonium lactate medium and fair growth with slight acidity in Fermi and Montesano's glycerol ammonium tartarate medium; produced good growth and acid without gas from arabinose, galactose, levulose, rhamnose, xylose, dextrose, lactose, maltose, sucrose, starch, mannitol ; slight to fair growth in dulcitol, glycerol, raffinose ; alkali in acetic, citric, lactic acids and no growth in salicin, cellulose and benzoic, formic, oxalic, salycilic, tannic, tartaric acids ; utilised nitrogen from ammonium citrate, ammonium dihydrogen phosphate, ammonium nitrate, ammonium oxalate, ammonium sulphate, ammonium tartarate, magnesium nitrate, potassium nitrate, sodium nitrate, alanine, asparagine, aspartic acid, creatin, creatinine, cystine, guanidine hydrochloride, glycine, glucocyamine, leucine, methionine, norvaline, ornithine hydrobromide, phenyl alamine, serine, threonine, tryptophane, tyrosine, urea and valine in presence of dextrose while glutamic acid and proteose peptone supplied both the carbon and nitrogen for growth and nitrates of Ag, Ba, Ca, Cd, Cu and Zn proved olygodynamic or toxic.

Fig. 1—Leaves of *Lochnera pusilla* showing disease symptoms

Host Range

When the host range of related and unrelated plants, especially on which bacterial disease is described from this laboratory was tried, it was observed that the pathogen is specific to *Lochnera pusilla* only and did not infect *Aegle marmelos* Corr., *Allamanda grandiflora* Lam., *Alysicarpus rugosus*, DC., *A. vaginalis* DC., *Amaranthus viridis* L., *Beaumontia grandiflora* Wall., *Begonia* sp., *Brassica oleracea* L., *Bridelia hamiltoniana* Wall., *Butea frondosa* Konig., *Caesalpinia sepiaria* Roxb., *Capsicum annuum* L., *Cajanus cajan* Millsp., *Carissa carandas* L., *Cassia tora* L., *Citrus aurantifolia* Sw., *Clerodendron phlomoides* L., *Cyamopsis tetragonoloba* (L.) Taub., *Desmodium diffusum* DC., *D. gangeticum* DC., *Dolichos lablab* L., *Ervatamia coronaria* Stapf., *Erythrina indica* Lam., *Euphorbia pulcherrima* Willd., *Gossypium herbaceum* L., *Ipomoea muricata* R. & Sch., *Lawsonia alba* Lam., *Lochnera rosea* Reichb., *Medicago sativa* L., *Melilotus indica* All., *Nerium indicum* Mill., *Phaseolus coccineus* Lam., *P. lunatus* L., *P. vulgaris* L., *Piper betle* L., *Pisum sativum* L., *Plumeria acutifolia* Poir., *P. rubra* L., *Ricinus communis* L., *Sesbania aegyptiaca* Prain., *Soja Max* (L.) Piper, *Stizolobium deeringianum* Bort., *Tabernaemontana dichotoma* Roxb., *Tamarindus indica* L., *Tectona grandis* L., *Tephrosia purpurea* Pers., *Thevetia nerifolia* Juss., *Trichodesma zeylanicum* R & Br., *Trigonella foenum-graecum* L., *Vallaris solanacea* O. Ktze., *Vigna sinensis* Endl., and *Xanthium strumarium* L.

Since there is no record of a *Xanthomonas* species on Apocynaceae and as the pathogen is highly specific to its suscept, it is proposed to designate it *Xanthomonas lochnerae*. sp. nov.

Xanthomonas lochnerae Patel, Thirumalachar and Bhatt sp. nov. On leaves of *Lochnera pusilla* Schum. Found at Poona in June, 1954. Leg. M. J. Thirumalachar.

Short rods ; single polar flagellum ; Gram negative ; capsulated ; non-spore former ; agar colonies, smooth, circular, entire, raised, butyrous and yellow ; gelatin liquefied ; starch hydrolysed ; casein digested ; milk peptonised and litmus reduced ; hydrogen sulphide and ammonia produced from peptone ; nitrite and ammonia not produced from nitrate ; good growth in synthetic nitrate and Czapek's media ; acid without gas from arabinose, dextrose, lactose, sucrose and starch ; no growth in salicin ; optimum temperature for growth 27°-30°C.; thermal death point about 51°C.

Summary

A new bacterial leaf-spot disease of *L. pusilla* is reported for the first time and detailed morphological, cultural, biochemical and host responses of the pathogen are given. The pathogen is designated as *Xanthomonas lochnerae* on account of its host specificity.

Disease-specimens and pathogenic culture are deposited in the herbarium and the culture collection of the Plant Pathologist to Government, Bombay State, College of Agriculture, Poona, India.

The culture is also deposited with Dr. M. P. Starr, Department of Bacteriology, University of California, Davis. California, U S. A.

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SOME STUDIES ON A VIRULENT STRAIN OF *CLADOSPORIUM HERBARUM* (Link) Fr. ON WHEAT

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(Accepted for publication, October 24, 1955)

A severe attack of *Cladosporium* on the wheat crop was reported in the year 1953 from the low-lying tracts of Choupasni Dam area near Jodhpur, where the crop was growing under moist and shady conditions.

Brooks and Hansford (1923) have described *Cladosporium* on wheat to be a common and mild parasite affecting dead or half-dead plant tissues. Bennett (1928) has attributed the attack of *Cladosporium* as secondary to *Ophiobolus graminis* Sacc., and *Erysiphe graminis tritici* El. Marchal. Bongini, Sibillia and Voglino (1927) and Fraser (1932) in South Wales have recorded *Cladosporium* as one of the organisms causing sooty moulds of wheat. Bockmann (1933) working with blackening fungi of cereals found *Cladosporium* to be an active parasite. Ogilvie (1943) in Britain has reported reduction in yield due to the attack of the fungus on ear heads. Moore (1948), Blair and Morrison (1949), and Nilsson (1950) have included *Cladosporium herbarum* (Link) Fr. as one of the fungi affecting wheat in association with some other fungus or insect pest. Nance and Nellie (1950) have reported *Cladosporium herbarum* (Link) Fr. causing damage to the wheat in Kentucky, U. S. A.

The fungus has not been reported so far from India as a pathogen. The present paper forms the subject matter of some studies on the various aspects of the fungus and the disease it causes.

Symptoms of the disease—Bennett (1928) has described the symptoms on wheat in detail. According to him, the disease appears on the ear heads in moist localities and produces a greenish black mouldy growth on the affected parts. He, however, did not find the disease to be severe on leaves and stems of wheat.

In the case of the disease under investigation, observations on the symptoms were recorded in the second week of February 1953, when the crop was nearly three months old. The fungus was found to infect leaves rather severely. Besides the development of a greenish black mouldy growth on the affected surface as the usual symptom of the disease, chlorotic patches also developed on the other side of affected areas.

Material and Methods

Infected leaves of the local wheat variety were collected. The fungus was isolated by the usual methods and incubated at 20°C. All the isolates yielded the same fungus, which was purified by taking

single spore cultures. For testing the pathogenicity of the fungus the local wheat variety was used.

An authentic culture of *Cladosporium herbarum* (Link) Fr. on wheat was obtained for comparative studies from the Indian type culture collection of fungi maintained in the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

Conidia from fifteen-day-old cultures were used for cultural, germination and infection studies. Inoculations were made by smearing the conidia on the leaves of the plants grown in pots.

These experiments were always conducted in triplicates and the average of the readings was taken into consideration. The colony diameter of cultures was measured in millimetres taking an average of two diameters at right angles to each other.

Experimental

Pathogenicity—The leaves of seven-day-old plants were smeared with a conidial mass. Inoculated plants were kept under 100 per cent humidity for forty-eight hours and thereafter were removed to glass cages having adequate air supply. The fungus started developing on the leaf surface on the 3rd day of inoculation. The experiment was repeated three times to confirm the results. The culture obtained from the Indian Agricultural Research Institute, New Delhi, however, did not infect the inoculated leaves.

Influence of Temperature on the Intensity of Infection- The influence of temperature on the intensity of infection was studied by making inoculations at intervals during the period from October, 1953 to March, 1954. The results obtained with inoculations made from 30th December, 1953, to 15th February, 1954, were positive, when the temperature conditions of the glass chamber varied from 41° to 72°F. This shows that the low temperatures prevailing during winter months play an important part in infecting the crop.

Effect of Sugar Solution as a spray on the Intensity of Infection— Inoculations were made as usual on two sets of wheat seedlings. One set of plants was atomized with sterilized water and the other with two per cent sterilized sucrose solution. The temperature of the glass cages varied from 41° to 72°F. It is evident from the results presented in table 1 that two per cent sucrose solution exercises a stimulatory effect on the activity of the fungus resulting in more intense infection. Intensity of infection is indicated in terms of + and ++, the former indicating the greenish mouldy growth of the fungus only in specks, scattered over the leaf surface, and the latter indicating the formation of bigger spots of the mouldy growth.

TABLE 1

The effect of sugar solution as a spray on the intensity of infection

	Date of Inoculation.	Temperature		Intensity of infection.	Percentage infection.	Remarks.
		Minimum °F.	Maximum °F.			
Sterilized water	31.1.54	68	72.0.	+	40	Observations were recorded on the 5th day after inoculation.
	15.2.54	68	72.5.	+	45	
Two per cent sucrose solution.	31.1.54	68	72.0.	+ +	80	
	15.2.54	68	72.5.	+ +	88	

Humidity was observed to have a profound effect on the intensity of infection under optimum conditions of temperature. A minimum exposure of 24 hours was necessary for obtaining infection, but a prolonged exposure to 100 percent humidity for 72 hours or more in the glass cages, though increased the intensity of infection, adversely effected the health of the plants.

TABLE 2

The role of insects in increasing the intensity of infection

Treatment	Intensity of infection	Percentage of plants showing infection	Remarks
Inoculation in the presence of insects.	++	78	The experiment was repeated thrice and the mean was taken.
Inoculation in the absence of insects.	+	38	

Insects in relation to infection: It was observed that certain insects were associated with infection. A number of insects belonging to the species *Aphis brassiceae* L. were collected and fed on wheat seedlings and flowering twigs of *Brassica campestris* Linn. under a glass chimney cover for one week. Later on, inoculation with the fungus was made and the insects were liberated on the plants. Simultaneously, another set of wheat plants was also inoculated with

the same fungus but without insects. It was found that after sometime insects had deposited some secretion on the leaves in the form of small chocolate-coloured dots, which effected the severity of infection. In table 2 are given the results which indicate the role of insects in increasing the intensity of infection.

Fungus Morphology

Histopathological observations.—The infection histology of the fungus has been studied by cutting free-hand sections of the leaves on the 3rd day of inoculations. The germ tube enters the host either through stomata or directly by rupturing the epidermis (fig. 1, a, b). There is no appressorial formation, but the germ tube is sufficiently pointed, as it penetrates the epidermis. The mycelium remains confined only to the epidermal layer in an intercellular or intracellular position, and does not seem to form any haustoria (fig. 1 c, d). Similar observations were made by Bond (1938) in the case of *Cladosporium fulvum* Cooke.

Mycelium.—The vegetative mycelium is greenish brown, septate and sparsely branched. The older hyphae are comparatively more closely septate. The cells of the hyphae are filled with coarse, granular protoplasm and numerous oil globules. The mycelium forms a thick weft during growth. Measurements of hyphae vary from 3 to 4 μ when grown on Oat meal agar; there is little or no distinction between the vegetative and reproductive mycelia.

Conidiophores.—The old vegetative hyphae in the centre of the colony begin to produce conidiophores on the third day of inoculation. They frequently get irregularly branched, become closely septate and acquire olive brown colour.

Conidia.—Brooks and Hansford (1923) have described the morphology of the conidia of *Cladosporium herbarum* (Link) Fr. in detail. Their observations were more or less similar to ours. Conidia originate in chains, both terminally and laterally, on the conidiophores, and their development is in acropetal succession. The basal conidia are most frequently uniseptate and the terminal ones are nonseptate. The usual number of conidia in a single chain varies from 4 to 6 (fig. 2 b, c). The shape of conidia is oval or round, and the colour of the older ones is greenish brown and that of the younger ones is much lighter. They measure 6×5 μ. The fungus obtained from Indian Agricultural Research Institute was found to be similar to this. (fig. 2 a, b, c).

Cultural characters—The local fungus shows some variation from the Delhi isolate on Oat meal agar, viz., a lighter colour of the colony and a more fluffy and rapid growth than those of the latter. Temperature range for growth in the case of both the isolates is from 10°C to 32°C. Both the isolates could withstand a wide range of pH (3.0—9.0). The optimum lies between pH 4.5 to 6.0. Sugar in varying concentrations in Richards synthetic agar (Riker and Riker, 1936) has been found to have a stimulatory effect on the growth of

the fungus. Optimum growth is obtained at a concentration varying from 10 to 15 per cent.

Discussion

Saccardo (1886) has recognized a number of varieties of *Cladosporium herbarum* (Link) Fr. This distinction is based on morphological characters and the relationship between the host and the parasite.

The two isolates, *Cladosporium herbarum* (Link) Fr. from the Indian Agricultural Research Institute and the fungus under study do not show any differences in respect of morphology. However, certain differences in their cultural characters have been recorded. They are (a) colour of colony, (b) type of growth and (c) the rate of growth.

A difference in the pathogenicity of the two isolates has been noticed. Since facultative parasites vary greatly in their degree of virulence, which is affected by various factors, the present fungus is identified only as a virulent strain of *Cladosporium herbarum* (Link) Fr.

The results obtained have also shown that sugar stimulates the fungal growth and improves it's infectivity. Since the secretion given out by the insects is also of sugary nature according to T. V. Ran: Krishna Ayar (1931), the role of insects becomes secondary in importance, and the disease is primarily caused by the fungus whose activity is increased by the sugary secretions of the insects,

Summary

The paper deals with a virulent strain of *Cladosporium herbarum* (Link) Fr., causing sooty mould of wheat in Jodhpur. The most important symptom of the disease is the greenish black mouldy growth of the fungus on the leaves.

The following factors were found to have a profound effect on the pathogenicity of the fungus.

(*a*) A minimum period of 24 hours under conditions of saturated atmosphere is needed for successful infection.

(*b*) Low temperatures prevailing during winter (41°F to 72°F) increase the intensity of infection.

(*c*) Two per cent sugar solution aggravates the virulence of the fungus.

(*d*) Association of the fungus with *Aphis brassiceae* L. results in more severe infection.

Morphology and histology of the fungus have been studied. Some differences were noticed in the two isolates in artificial culture.

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Explanatiom of Text Figures

Figure 1

a. Penetration of the germ tube through a stomata.
b. The same directly through the epidermis.
c. Intracellular mycelia without haustoria.
d. Intracellular mycelia.

Figure 2

a. Germinating conidia of the fungus under study.
b. Conidiophore with conidia attached to the fungus under study.
c. Conidiophore with conidia attached.
d. Germinating conidia of *Cladosporium herbarum* (Link) Fr. obtained from I.A.R.I., New Delhi.

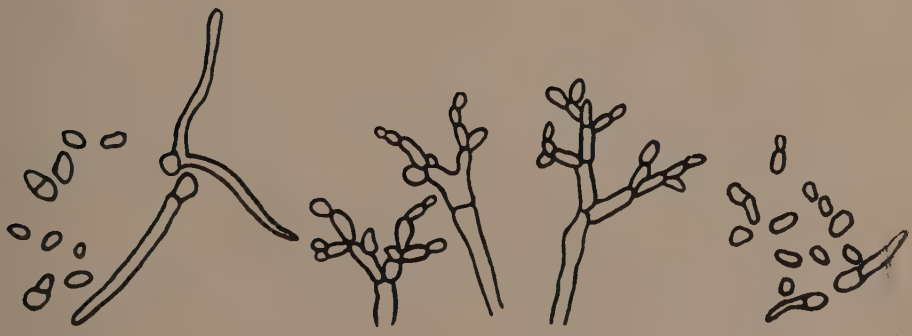
* Original not seen.

1. a.

1. c.

1. b.

1. d.



2. a. 2. b. 2. c. 2. d.

NOTES ON MISCELLANEOUS INDIAN FUNGI II*

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19. *Erysiphe polygoni* DC. in *Flor. Fr.* 2, p. 272, 1805; Saccardo, *Sylloge Fung.* 1, p. 19, 1878; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, 1, p. 22, 1931.

Spots amphigenous; mycelium effuse, persistent; *Perithecia* superficial, loose, globose, scattered, rarely gregarious, yellow to start with, then red and finally dark red, measuring 108—162×97—155 μ. *Tendrils* (appendages) few or many, number varying from 5 to 12, hyaline, unbranched, generally curved, rarely straight, about 7 μ thick. *Asci* usually 7—8 in each perithecium, hyaline or sub-hyaline, clavate-oblong, shortly stipitate, wall uniformly thick, 48—72×30—41 μ; *Ascospores* one to many (1—6 seen), sub-hyaline with granular protoplasmic contents, sometimes one end tapering, other round, pear shaped, oval or oblong, measuring 22—30×11—18 μ (mostly 24—26×16 μ).

On living leaves, stems and pods of *Tephrosia purpurea* Pers., Upper Ridge Road, New Delhi, 15-1-1949 (R. L. Munjal); on living leaves of *Cassia laevigata* Willd., Lake area, Kodaikanal, Pulney Hills (South India) 19-12-1951 (R. L. Munjal) and on living leaves of *Vicia ervilia* Willd., Botanical area, Indian Agricultural Research Institute, New Delhi, 8-4-1950 (Girdhari Lal).

20. *Diatrypella borassi* sp. nov. (Fig. 1).

Perithecia immersed in stroma, later erumpent. Stromata linear, often running together, 2–3 mm. long and 1-1.5 mm. broad, 3-7 perithecia in one stroma with long protruding necks and opening by rupturing the epidermis. Perithecia embeded in cortex tissue, placed in one layer (monostichus), flask shaped but irregular in outline due to pressure of one another, dark brown in colour with olivaceous contents and measure 200—450μ in diameter; neck 200—500μ long. *Asci* innumerable, clavate, acute at apex and broader in the middle, with long drawn out stalk, hyaline, polysporus, paraphysate, measuring 108—154 ×11—16μ (mostly 125—140×12—14μ). *Paraphyses* few, multi-septate, hyaline, clavate-cylindrical, unbranched, 110—160×2—3μ. *Ascospores* allantoid, sub-hyaline, ends obtuse, olivaceous in mass, measuring 6—7×2μ and provided with two round vacuoles, one at each end.

On dead leaf stalks of *Borassus flabellifer* L., Flowerdale, Simla, 18.11.1948 (R. L. Munjal). Type specimen deposited at Herb. Crypt. Ind. Orient., Indian Agricultural Research Institute, New Delhi.

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*No. I of the series, giving description of 18 fungi, appeared in Indian Phytopathology **3**, pp. 105-116, 1950.

Perithecia immersa in stromata, postea erumpentia. Stromata linearia, saepe confluentia, 2—3 mm. longa, 1—1.5 mm. lata, 3—7 in singulis stromatibus, ornata collo longo protrudente atque patentia post rupturam epidermidis. Perithecia immersa in textus corticales, monosticha, urceolata sed ambitu irregularia ob mutuam pressionem, colore fusce brunnea, contentis olivaceis, magnitud. 200–450μ diam.; colla 200—500μ longa. Asci innumerabiles, clavati, acuti ad apicem, latiores ad medium, pedicello longe elongato praediti, hyalini, polyspori, paraphysati, magnit. 108—154μ (ut plurimum 125—140×12—14μ). Paraphyses rarae, multiseptatae, hyaline, clavato-cylindricae, haud ramosae, 110–160×2—3μ. Ascosporae allantoideae, subhyalinae, apicibus obtusis, olivaceae si congestae, magnit. 6—7×2μ atque praeditae duplici vacuolo rotundato ad utrumque apicem.

Typus lectus in petiolis emortuis *Borassi flabelliferi* L. in loco Flowerdale prope Simla, die 18 mensis novembris anni 1948 a R. L. Munjal. et positus in Herb. Crypt. Ind. Orient., in Ind. Agric. Res. Inst., New Delhi.

21. *Diatrypella quercina* (Pers.) Nits. in *Pyr. Germ.* p. 71, 1867 ; Saccardo, *Sylloge Fung.* I, p. 206, 1882.

Several perithecia embedded in a stroma (5 to 15 observed) uni or biseriate, mostly biseriate. *Stromata* pulvinate, at first embedded in the cortical tissue, later erumpent, dark brown with minute dots (pores) at the surface, 2–3 mm. in length and 1—1.5 mm. broad. *Perithecia* flask-shaped with rather swollen ostiole lined on the inside with innumerable sulcate periphyses ; basal portion globose, but sometimes angular due to the pressure of one another, measuring 500—790×400—530 μ in diameter, and the neck measuring 280—310μ and ostiole 100 to 110μ. *Asci* are fusoid, 52–95×8—12 μ, polysporus and narrowly pointed at the base ; *Ascospores* allantoid, strongly curved with blunt ends, olivaceous in colour and 7—12×2—3μ.

On dead twigs of *Quercus* sp., Bhowali, U. P., April, 1949 (K. M. Dutt).

22. *Glomerella alstoniae* sp. nov. (Fig. 2).

Perithecia embedded in cortex (bark tissue), stromate, 3—7 seen in a single stroma, uniseriate, subepidermal, globose or subglobose but sometimes irregular in outline due to pressure of one another, with a short neck, ostiolate, wall parenchymatous many layered thick, dark brown in colour and measure 120—140×170—220 μ ; *Asci* hyaline, sub-clavate to cylindrical, thin walled, 8 spored and measure 95—100×19—21 μ. *Ascospores* are hyaline, single-celled, oblong to ovate, straight or slightly curved, both ends rounded, uniseriate or biseriate and measure 18—20×5—10 μ. No paraphyses seen but periphyses present.

On dead twigs of *Alstonia scholaris* Br., Indian Agricultural Research Institute, New Delhi, 31.7.1949 (R. L. Munjal). Type specimen deposited in Herb. Crypt. Ind. Orient. I.A.R.I., New Delhi.

Perithecia immersa in textus corticales, 3—7 in singulis stromatibus, uniseriate, subepidermalia, globosa vel subglobosa, sed non raro irregularia ambitu ob mutuam pressionem, ornata collo brevi, ostiolate, parietibus parenchymaticis, multi-seriatis crassis, fusce brunnea colore, magnitud. 120—140×170—220μ. Asci hyalini, subclavati vel cylindrici, tenuibus parietibus praediti, octospori, magnitud, 95—100× 19—21μ. Ascosporae hyalinae, unicellulatae, oblongae vel ovatae, rectae vel tenuiter curvatae, apicibus rotundis, uniseriate vel biseriate, magnitud. 18—20×5—10 μ. Paraphyses nullae, sed periphyses visae.

Typus lectus in ramis emortuis *Alstomae scholaris* Br. in loco Ind. Agric. Res. Inst., in urbe New Delhi, die 31 julii,, anni 1949, a R. L. Munjal, et positus in Herb. Crypt. Ind. Orient.

23. *Rosellinia erianthi* sp. nov. (Fig. 3).

Perithecia superficial, almost black, single or gregarious, seated on subicle, globose to subglobose measuring 1-1.5 mm. in diameter, papillate, pore prominent ; wall hard, many layered. *Subicle* cottony in texture naples yellow in colour; *Asci* clavate with sub-acute apex, broader in the middle and tapering towards lower end, measure 65—75×10μ and have a hyaline wall ; *Ascospores* 8 in each ascus, uniseriate, boat-shaped, at first hyaline, later raw amber to mummy brown, 9—11.5×5 μ (mostly 10—10.5×5μ), provided with globular oil droplets, 3—4 μ in diameter, giving an appearance of psuedo-septum in some cases.

In collar region of dead clumps of *Erianthus munja* (=*Saccharum munja* Roxb.), Mycological area. Indian Agric. Res. Institute, New Delhi 29-12-1950 (B. L. Chona). Type specimen deposited at Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.

Perithecia superficialia, fere nigra, singula vel gregaria, subiculo innixa, globosa vel subglobosa, magnit. 1—1.5 mm. diam., papillata, poro prominente, parietibus duris, multi-seriatis. Subiculum gossypinum texture, colore "Naples Yellow". Asci clavati, apice subacuto, latiores ad medium, fastigati ad apicem inferiorem, magnitud. 65—75×10μ, parietibus hyalinis. Ascosporae 8 in singulis ascis, uniseriate, naviculares, primo hyaline, tum "Raw Amber" vel "Mummy Brown", unicellulatae, 9—11.5×5μ (ut plurimum 10—10.5× 5μ) ornatae globulis olei 3—4μ diam., qui monnumquam falsum septum simulant.

Typus lectus ad collum plantarum mortuarum *Erianthi munjae* (=*Sacchari munjae*), in regione mycologica in Inst. Agric. Res. in urbe New Delhi, die 29 mensis decembris anni 1950 a B. L. Chona, et positus in Herb. Crypt. Ind. Orient., in Ind. Agric. Res. Inst., New Delhi.

24. *Mycosphaerella smilacicola* (Schw.) Chona & Munjal comb. nov.

Syn. *Sphaerella smilacicola* (Schw.) Cke. in Saccardo, *Syll. Fung. I*, p. 524, 1882.

Depasea smilacicola Schw. in Grev.

Spots numerous, at first circular to oval, then irregular, mostly separate, rarely coalescing, light sorghum brown to avellaneous, with dark red, raised margins, mostly 4–12×3—8 mm. *Perithecia* dot-like, minute, innate, then erumpent, subepidermal, separate, epiphyllous, rarely hypophyllous, globose 80—120μ in diameter, ostiolate ; ostiole prominent. *Asci* club-shaped, shortly stalked, 36—48×8—12μ, hyaline paraphyses or paraphysoids absent, 8 spored, in two rows ; *Ascospores* hyaline, oblong, both ends round, somewhat tapering towards distal end, bicelled, 12—16×3—4μ, not constricted at septum.

On living leaves of *Smilax aspera* L., Nainital (U. P.), July, 1950 (R. Prasada).

There is a lot of confusion in literature about the use of the generic name *Sphaerella* (Fr.) Rabenhorst (1856) used for a fungus and *Sphaerella* Sommerf. (1824) having been used earlier for an alga. Both the names continue to be used by Mycologists and Algologists and this results in unnecessary confusion. Johanson (1884), therefore, proposed the name *Mycosphaerella* to accomodate the fungus genus *Sphaerella*. Since then a number of species have been transferred from *Sphaerella* to *Mycosphaerella* but some mycologists have persisted in the use of *Sphaerella* and even recommended its conservation. But we feel that to avoid any further confusion. *Mycosphaerella* should be preferred and this new combination is, therefore, proposed.

25. *Cerotelium fici* Arthur in *Bull. Torrey Bot. Club*, **44** : 509, 1917.

Uredo-sori hypophyllous, scattered ; roundish but sometimes irregular, mostly single, rarely confluent, cinnamon buff in colour, cushion-like ; subepidermal ; and measure 2—2.5 mm. in diameter ; *Uredospores* obovate-globoid, 18—25×16—20μ, mostly 21—24×16—18μ, hyaline, with a sparsely echinulate wall, 1-1.5 μ thick.

Infecting leaves of *Ficus infectoria* Roxb., Indian Agric. Res. Institute, New Delhi, 18-11-1948 (R. L. Munjal).

26. *Hyalopsora orientalis* sp. nov. (Fig. 4 a and b).

Uredo-sori hypophyllous, subepidermal, scattered on yellowish irregular, indefinite spots, pustular, pulverulent, about 1 mm. in diameter, numerous, ochre to ochre brown in colour, covered by a thin peridium ; *Uredospores* are of one type, golden yellow in colour, polygonal, ellipsoid, ovoid or obovoid, very shortly stalked, 30—35×56—63μ (mostly 34—35×58—60μ), stalk hyaline and narrowly pointed; *Epispore* hyaline, thick (thickness not uniform), mostly 5—6μ and provided with 4 equatorial germpores. Teleuto-sori diffuse, intracellular, *Teleutospores* 4—30 celled, cruciate, olivaceous yellow changing to dark brown and then subopaque, 22—23×16—17μ. Spore wall dark brown, smooth.

On living leaves of *Adiantum* sp., Kufri, Simla Hills, 18-5-1949 (M. L. Gattani). Type specimen deposited at Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.

Hyalopsora orientalis spec. nov.

Uredo-sori hypophylli, subepidermales, dispersi in maculas indefinites, luteas, irregulares, pustulares, pulverulenti, ca. 1 mm. diam., plurimi, ochracei vel ochraceo-brunnei colore, operti peridio tenui. Uredosporae uniformes, aureo-luteae colore; polygonales, ellipsoideae, ovoideae vel obovoideae, brevissime pedicellatae, 30—35×56—63μ (ut plurimum 34—35×58—60μ), pedicello hyalino atque anguste acuto. Episporium hyalinum, crassum (sed non uniformiter) ut plurimum 5—6μ atque ornatum 4 germinationis poris. Teleutosori diffusi, intracellulares, Teleutosporae 4—30-cellulati, cruciati, olivaceo—lutei, vergentes in brunneum fuscum atque tandem subopaci, 22—23×16—17μ. Sporarum parietes fusce brunnei, leves.

Typus lectus in foliis viventibus *Adianti* spec. and Kufri in montibus prope Simla, die 18 maii, anni 1949 a M. L. Gattani, et positus in Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.
Hyalopsora adianti-capilli-veneris Syd. recorded on *Adiantum capilli-veneris* differs from the above in having smaller spores.

27. *Puccinia tweediana* (Speg.) Ramakrishnan T. S. & K. in *Proc. Ind. Acad. Sci., Sect.* B, **28**, pp. 62-63, 1948.

Spots almost circular on leaves, amphigenous, mostly epiphyllous yellowish, later dark brown, 4—7 mm. in diameter ; on stems fusiform to elliptic 5—8×2.5—3 mm. ; *Pycnia* not known ; *Aecia* hypophyllous, filling up the entire spot, cup shaped, cups prominent, with raised peridia, orange coloured when fresh, rust coloured when dry ; *Aeciospores* in chains, subglobose, light yellow, wall very finely verrucose, mostly 18—20×17μ. *Uredo-sori* absent. *Teleuto-sori* subepidermal, mixed with aecia or surrounding aecial cups, hypophyllous, very rarely epiphyllous, appearing at first as chocolate coloured raised pustules, but later turning black and encrusted. Black crusts specially prominent on stem, sometimes completely girdling the stem. Teleutospores bicelled, slightly constricted at the septum, clavate, apex obtuse, thickened upto 10 μ, light yellowish to yellowish brown depending on the maturity of the spores, dark brown in mass, 39—59.5×14—21μ, mostly 49×17μ. Stalk hyaline, long, slightly thickened at the junction of spore, tapering below.

On living leaves, stem and petioles of *Dicliptera bupleuroides* Nees, Flowerdale, Simla 17-11-1948 (R. L. Munjal).

Telial stage has not been recorded previously on this host. Ramakrishnan, T. S. and K. reported the telia of this fungus to be scanty but under Simla conditions and on this host species, these are formed in plenty.

28. *Ganoderma lucidum* (Leys.) Karst in Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1**, p. 98, 1931. forma *naiae* forma nov.

Fruiting body hard, shining, stalked, stalk and upper surface of pileus of the same colour with slightly different shades at places.

Young fructification resembling a cobra while the mature one resembling a hooded cobra. *Pileus convex* ; upper surface woody, carob brown with a thin rim, hay's russet in colour at the exterior ; 27×9 mm. to 35×16 mm. Lower surface light buff to light ochraceous buff in colour. Hymenium not uniformly thick ; pores round, tubular, in a single layer. *Stipe* lateral, oblong, straight or slightly curved, roundish, wrinkled or slightly flattened at places, 50—90mm. in length and 4—9mm. in diameter. *Spores* light orange yellow to xanthine-orange ; one celled, ovate, with verrucose wall and hyaline, papillate apex, 7—11×5—7μ.

In dead humus on ground ; Christian Lodge Simla, July,1949 (L. M. Joshi).

Differs from other known forms in its typical cobra-shaped appearance, found growing in nature.

Ganoderma lucidum Karst f. *naiae*, forma nova.

Ab aliis formis hucusque notis differt forma typica Naiae tripudiantis.

29. *Phoma sirodesmii* sp. nov. (Fig. 6).

Pycnidia superficial, subglobose to ovate, ostiolate, light brown to dark brown in colour with a thin parenchymatous wall and measure 40—60×36—45μ (mostly 55—60×40—45μ). *Pycnospores* hyaline, elliptic to ovate, single celled, rounded at both ends, with one oil globule, measuring 6—10×5—7μ (mostly 7—8×6μ) and are borne on short elliptic hyaline cells arising from the pycnidial wall.

Developing on the juice secreted by *Laccifer lacca* Kerr parasitising *Ficus infectoria* Roxb., Indian Agricultural Research Institute, New Delhi, 5-1-1951 (R. L. Munjal). Type specimen deposited at Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.

Phoma sirodesmii spec. nov.

Pycnidia superficialia, subglobosa vel ovata, ostiolata, pallide vel fusce brunnea colore, parietibus tenuibus parenchymaticisque, magnit. 40—60×36—45μ (ut plurimum 55—60×40—45μ). Pycnospoiae hyaline, ellipticae vel ovatae, unicellulatae rotundatae ad utrumque apicem ornatae unico globule olei, magnitud. 6—10×5—7μ (ut plurimum 7—8×6μ), insidentes cellulis brevibus ellipticis hyalinis quae ex parietibus pycni-dialibus oriuntur.

Typus enutritur suco producto a *Laccifero lacca* Kerr. qui parasitice incolit *Ficum infectorium* Roxb., et lectus in Ind. Agric. Res. Inst,, in urbe New Delhi die 5 mensis januarii anni 1951 a R. L. Munjal, et positus in Herb. Crypt. Ind. Orient.

Some of the *Ficus infectoria* trees which are growing at the I.A.R.I., New Delhi on both sides of the roads as ornamental plants, showed infection of twigs by a lac insect, identified by the Head of

the Division of Entomology, I.A.R.I., New Delhi as *Laccifer lacca* Kerr. After sometime, a black sooty mould was found developing on these trees and the spread was specially rapid after rains, covering the entire surface of leaf, mostly upper (rarely both) as a thin mat, also extending to the twigs, leaf petioles etc., giving the whole tree a charred appearance. The affected leaves turn yellow and then red and fall off prematurely. Microscopic examination of the fungus proved it be a dematiaceous fungus with very scanty creeping olivaceous mycelium but with copious formation of phaeodictyospores borne in chains mixed with pycnidia. The dictyospores are olivaceous brown to dark brown and of variable size. Proliferating end cells give rise to new spores, thus bearing the spores in chains, while some of the young conidia, instead of cutting bigger cells of another conidium give rise to pycnidium. With the cessation of rains and the drier period that follows the pycnidial formation is arrested but dictyospores continue to be formed. Infected branches of the tree perpetuate the disease. Spores taken from the infected material from the tree and tested for germination in water from time to time, were found to be viable throughout the year. Pruning of the diseased branches has given encouraging results in controlling the disease. In no case sooty mould was found on a plant which was not affected by *Laccifer lacca.*

In literature, we find a fungus resembling in certain morphological features to the above described fungus under the name *Coniothecium scabrum* McAlpine (Mason, E. W. –Annotated account of Fungi received at Imperial Mycological Bureau, List II, 1933, illustrated by S. P. Wiltshire). McAlpine who originally described this fungus from Australia (Fungus Diseases of Citrus in Australia and their Treatment, Agr. Deptt. Victoria, Melbourne p. 80, 1899) clearly mentions that the mycelium is "scanty and creeping". The illustrations of the fungus both by McAlpine and Wiltshire, show the conidia as borne in chains. Even basing the identity of the fungus on the conidial stage, it should better be referred to *Sirodesmium* deNot. and not *Coniothecium* Corda. Again, although Dr. Wiltshire observed the Phoma stage, he did not see the spores being abstricted in the pycnidium. We feel that in the presence of a higher developmental stage of a fungus, it would be wrong to call it by the lower stage. We, therefore propose it as a new species of *Phoma* to accomodate this fungus.

30. *Phyllosticta grewiae* Died. apud Sydow & Butler in *Ann. Mycol.* **14**, p. 181, 19 6; Saccardo *Syll. Fung.* **25**, p. 75, 1931; Butler & Bisby in *Sci. Monogr. Ind. Coun. Agric. Res.* **1**, p. 161, 1931 (Fig. 7).

Spots circular, 3.5 to 9 mm. in diameter (mostly 6-8 mm.), amphigenous, scattered, sometimes coalescing and involving the whole leaf, purplish brown in colour, changing to dark brown, and with ash coloured centre on the upper surface. *Pycnidia* are hypophyllous, globose parenchymatous and many layered thick, scattered, numerous, light brown in colour and measure $90-120\times85-110\mu$ with a prominent ostiole. *Ostiole* measuring $15-23\mu$, rounded and having smaller and darker cells than those of the pycnidial wall. *Pycnos-*

pores are hyaline, single-celled, oblong, slightly tapering towards the distal end and measure 4—8×3μ (mostly 6—7×3μ); borne on hyaline, single-celled conidiophores with papillate apex measuring 10—13×4μ.

On living leaves of *Grewia asiatica* L., Entomological area, I.A.R.I., New Delhi, 27-7-1951 (R.L. Munjal).

Grewia asiatica is an important commercial fruit plant. The disease was found to cause considerable damage to foliage.

31. *Phyllosticta heterophragmae* sp. nov. (Fig. 8).

Spots are irregular in outline, at first 8×6 mm., then 18×14 mm. and later indeterminate, starting as single small spots but later coalescing, buff coloured with purplish margin. *Pycnidia* minute, scattered, mostly epiphyllous, dot-like, subepidermal, dark brown, 70—125×140 160μ, parenchymatous, seated in the palisade tissue; ostiolate, ostiole protruding to the surface and measures 20—25μ. *Conidiophores* hyaline pointed, 10—21μ (mostly 10 — 16 × 1.5 μ). *Spores* hyaline, single celled, oblong, lower end tapering, some fusiform, measure 6—8×1.5—2μ.

On living leaves of *Heterophragma adenophyllum* Seem., I.A.R.I., New Delhi March, 1949 (M. H. Rao) Type. Type deposited at Herb. Crypt. Ind. Orient., I. A. R. I., New Delhi.

Phyllosticta heterophragmae spec. nov.

Maculae irregulares ambitu, primo 8×6 mm., tum 18×14 mm., et tandem indeterminatae, initium ducentes ut maculae parvae, sed postea coalescentes, bubalinae marginibus purpureis. Pycnidia minuta, dispersa, ut plurimum epiphylla, puncti similia, subepidermalia, fusce brunnea, 70—125×140—160μ, parenchymatica, insidentia textibus vallaribus, ostiolata, ostiolo ad superficiem protruso, magnitud. 20—25μ. Conidiophori hyalini, acuti, 10—21μ (ut plurimum 10—16×1.5μ). Sporae hyalinae, unicellulatae, oblongae, fastigatae ad apicem inferiorem, nonnullae fusiformes, magnitud. 6—8×1.5—2μ.

Typus lectus in foliis viventibus *Heterophragmae adenophyllum* Seem. in loco I. A. R. I., in urbe New Delhi, mense martio anni 1949 a M. H. Rao. et positus in Herb. Crypt. Ind. Orient.

32. *Septoria adhatodae* sp. nov. (Fig. 9).

Spots amphigenous, dark brown with broad slightly raised, dark margins, irregular in outline, singly 5—10 mm. in diameter, usually confluent and forming larger spots covering several square inches. Older spots have ashy coloured centres. *Pycnidia* hypophyllous, dot-like, separate, subepidermal, thin-walled, globose, parenchymatous, dark brown in colour, 50—63μ in diameter (mostly 52—55μ), ostiolate; *Ostiole* broad, 11—15μ in diameter. *Pycnospores* hyaline, usually one septate, rarely 2 or 3 septate, variously curved, cylindrical, ends rounded or sometimes bluntly pointed, not constricted at the septum and measure 18—40×3.5μ.

On living leaves of *Adhatoda vasica* Nees, Ridge Road, New Delhi, 10-2-1951 (R. L. Munjal) Type. Type deposited at Herb. Crypt. Ind. Orient., I. A. R. I., New Delhi.

Septoria adhatodae spec. nov.

Maculae amphigenae, fusce brunneae, marginibus latis parumper elevatis, colore profundioribus, irregulares ambitu, 5—10 mm. diam., ut plurimum confluentes atque efformanates maculas largiores spatium monnulorum pollicum digitorum operientes. Maculae vetustiores cinereae ad centrum. Pycnidia hypophylla, puncti similia, separata, subepidermalia, tenuibus parietibus praedita, globosa, parenchymatica, fusce brunnea colore, 50—63μ diam., (ut plurimum 52—55μ), ostiolata. Ostiolum latum, 11—15μ diam. Pycnosporae hyalinae, ut plurimum uniseptatae, raro bis vel ter septatae, varie curvatae, cylindricae, apicibus rotundatis vel monnumquam hebetibus, haud constrictae ad septum, magnitudinis 18—40×3.4μ.

Typus lectus in foliis viventibus *Adhatodae vasicae* Nees in via Ridge Road vocata, in urbe New Delhi, die 10 februarii anni 1951, a R. L. Munjal, et positus in Herb. Crypt. Ind. Orient., I. A. R. I., New Delhi.

33. *Diplodia frumenti* Ell. & Ev. in *Journ. Myc.*, 1886, p. 103, Saccardo, *Syll. Fung.* **10** : 292, 1892.

Pycnidia globose, ostiolate, ostiole conical, often gregarious or in a linear series, formed just below the testa of the kernel ; *Pycnospores* ellipsoid, hyaline, thick walled, & single celled when young, measuring 16—22×11—14μ (mostly 18.6—20×11—12 μ). Mature spores bicelled, dark brown, not constricted at the septum and measure 17—19×11—12μ.

On grains of *Zea mays* L., Mycological area, I. A. R. I., New Delhi, October, 1948 (C. B. Sahaya).

The disease is rare in India and affects only a few mature grains in the cob, which are converted into a black sooty mass due to the aggregation of the hyphae of causal organism. The mycelium is septate, olivaceous, later dark brown, forms a stromatic mass on which pycnidia are formed. The perfect stage of this fungus has been reported by Eddins & Vorhees (*Phytopath.* **23** : 63—72, 1933) as *Physalospora zeicola* Ell. & Ev. but we have not come across the ascigerous stage of this fungus in India.

34. *Marssonina zanthoxyli* sp. nov. (Fig. 4 c).

Spots dark purple, more prominent on undersurface of leaves, 4—5.5 mm. in diameter, irregular in shape, scattered, later coalescing, elliptic on twigs ; *Acervuli* hypophyllous, maculicole, dot-like, scattered singly, at first subepidermal, later erumpent, pulvinate, purplish brown, 200—300μ in diameter. *Conidia* elliptic to club-shaped, curved, rounded above but tapering below, bicelled, hyaline and measure 19—31×4μ (mostly 21—24×4μ). *Conidiophores* are hyaline with pale green tinge, single celled, straight or slightly curved bearing conidia singly and measure 6—7×3μ.

On living leaves of *Zanthoxylum alatum* Roxb., Simla, 28-12-1950 (V.C. Lele) Type. Type deposited in Herb. Crypt. Ind. Orient., I. A. R. I., New Delhi.

Marssonina zanthoxyli spec. nov.

Maculae fusce purpureae, eminentiores in inferiore pagina foliorum, 4—5.5 mm., irregulares forma, dispersae, tandem coalescentes, ellipticae in ramulis. Acervuli hypophylli, maculicoli, puncti similes, dispersi, primo subepidermales, tandem erumpentes, pulvinati, purpureo-brunnei, 200-300μ diam. Conidia elliptica vel clavata, curvata, rotundata supra, sed fatigata infra, bicellulata, hyalina, magnit. 19—31×4μ (ut plurimum 21—24×4μ). Conidiophori hyalini sed pallide viride tincti, unicellulati, recti vel curvati tenuiter supportates conidia singula, magnit. 6—7×3μ.

Typus lectus in foliis viventibus *Zanthoxyli alati* Roxb., in loco Simla, die 28 decembris anni 1950 a V. C. Lele, et positus in Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.

35. *Hendersonia sabaleos* Ces. var. *phoenicis* Sacc. in Saccardo, *Syll Fung.* **10** : 326, 1892.

Spots elliptic or irregular, tawn coloured with broad dark brown margins on leaf stalk, axils or blades. *Pycnidia* at first subepidermal, then erumpent, separate, dot-like, round, 75—120μ in diameter, dark brown, wall many layered ; *Conidiophore* thin, smoky olive in colour bearing a single conidium at the apex ; spores oblong or cylindrical, mostly smoky grey, a few olive coloured, slightly curved, both ends rounded, usually 3-septate slightly constricted at the septa ; measuring 7—14×4—6μ (mostly 10—12×5μ). Immature spores measure 7—12×4—6μ (mostly 8—10×4—5μ).

In leaflets and rachis of *Phoenix dactylifera* L., Entomological area, I.A.R.I., New Delhi, 24-12-1949 (R. L. Munjal).

36. *Septocylindrium ranunculi* Peck. in 34th Rep. State Museum New York, p. 46, 1881; Saccardo, *Syll. Fung.* **4** : 223, 1886.

Spots numerous, amphigenous, roundish, sometimes irregular, whitish to light brown in colour and measure 1.5 to 3.5 mm. in diameter. *Conidiophores* emerging in tufts mostly on the undersurface of the leaf, measuring 20—35×2—3μ, hyaline, single celled ; *Conidia* hyaline, single celled, both ends rounded or one pointed, contents vacuolate and granular, 25—42×3—4μ (mostly 30—36×3μ).

On living leaves of *Ranunculus sceleratus* L., Khir Bhawani, Srinagar, 18-5-1950 (H. C. Arya).

37. *Attractium indica* sp. nov. (Fig. 4 d).

Sterile hyphae few, infesting scale insects, fertile hyphae prominent, hyaline, septate, grouped together in firm rows forming synnemata. *Synnemata* cylindric, slightly thickened at the base and swollen at the apex, bright orange coloured, sometimes branched, 3 to

5 mm. in length. *Conidia* hyaline, 6—13 septate, straight or curved, both ends acute, 80—136×6—8μ (mostly 100—130×6—7μ).

Parasitising scale insects on *Morus alba* L., Kalimpong, Darjeeling, July, 1949 (S. P. Raychaudhuri) Type. Type deposited in Herb. Crypt. Ind. Orient., I.A.R.I., New Delhi.

Attractium indicum spec. nov.

Hyphae steriles rarae, infestantes insecta squamaeformia, hyphae fertiles prominentes, hyalinae, septatae, simul acervate in lineas solidas efformantes synnemata. Synnemata cylindrica tenuiter crassa ad basim, atque tumescentia ad apicem, lucide aurantiaca colore, nonnumquam ramosa, 3—5 mm. longa. Conidia hyalina, 6—13-septa, recta vel tenuiter curvata, acuta ad utrumque apicem, 80—136×6—8μ (ut plurimum 100—130×6—7μ).

Typus lectus infestans insecta squamaeformia super *Morum albam* Linn. in loco Kalimpong prope Darjeeling, mense julio anni 1949 a S. P. Raychaudhuri et positum in Herb. Crypt. Ind. Orient., I. A. R. I., New Delhi.

38. *Arthrobotrym velutinum* Butler (Fig. 4 e).

"Late effusum, gregarium, velutinum, epiphyllum, atrum, stipitibus cylindraceis, glabris, 1—1¾ mm. altis, 30 - 60μ latis, ex hyphis fuligineis, septatis, 4—8μ latis compositis, conidiis capitatis, ex hypharum apice relaxato oriundis, obclavatis, dilute brunneis, 5 (rariuo pluri—) septatis, 60—70×12—18μ".

"In pagina superiore foliorum *Thysanolaenae acariferae* Nees (=*T. procera* Mez.) Dehra Dun, Indiae, legit Inayat Khan, Oct. 24, 1903."

Also collected on the same host at Sindai, Khasia hills, Assam Jan. 1916 (L. S. Subramaniam), at Sim's Park, Coonoor, Nilgiris, 16-12-1951 (R. L. Munjal). The leaves were attacked by scale insects.

A recent collection by the junior author showed it to be a different species than those recorded in literature but while referring to the herbarium specimens in Herb. Crypt. Ind. Orient., we found it to match exactly with *A. velutinum* Butler, in which the latin diagnosis of the fungus as made by Sir Edwin J. Butler on Jan. 16th 1904, also existed, which has been reproduced above. This species has not been effectively published by Sir Edwin, though the name occurs in the printed list of Mycological specimens at the Agri. Res. Inst., Pusa, May 1921 (published 1922).

Our sincere thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his keen interest, valuable criticism and encouragement. We are also indebted to Rev. Father Dr. H. Santapau, Head of the Biology Department, St. Xavier's College, Bombay for rendering the latin diagnosis of new species.

Division of Mycology and Plant Pathology,
Indian Agricultural Research Institute,
New Delhi.

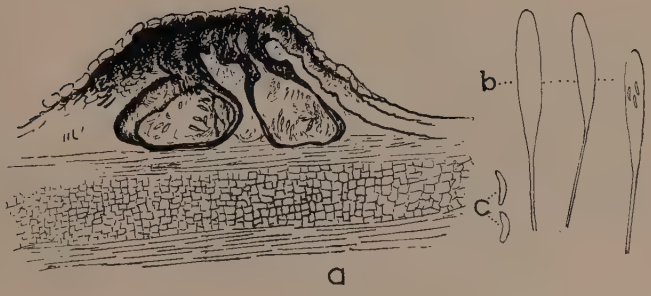

Fig. 1

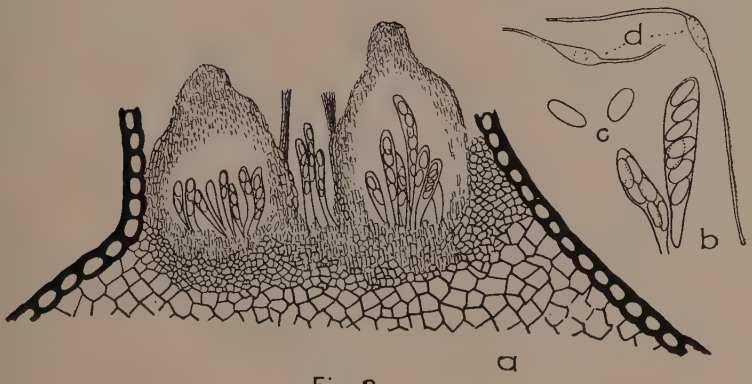

Fig. 2

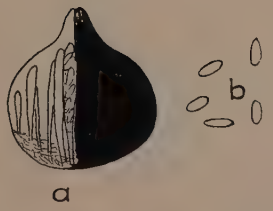

Fig. 3

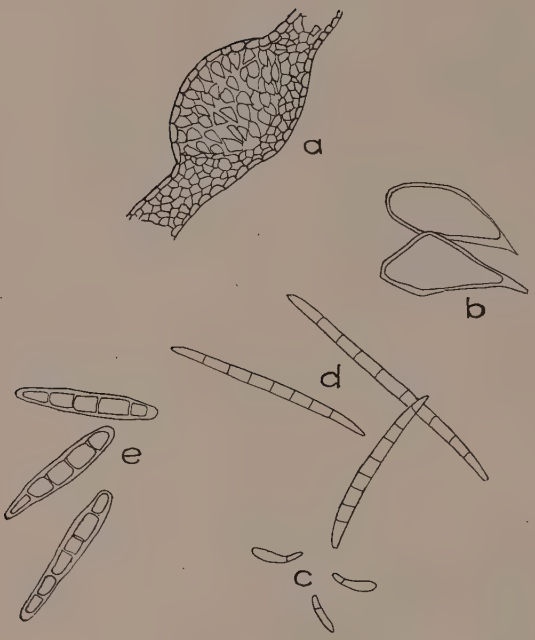

Fig. 4

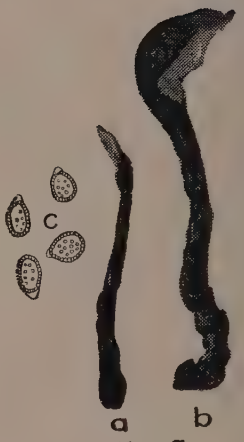

Fig 5

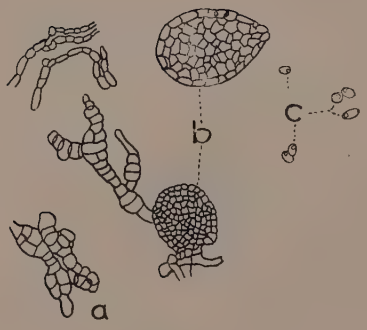

Fig. 6

Fig. 7

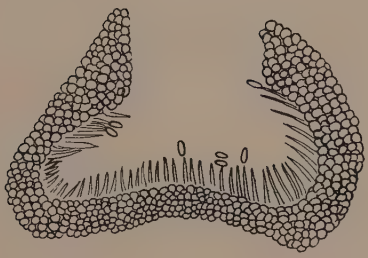

Fig. 8

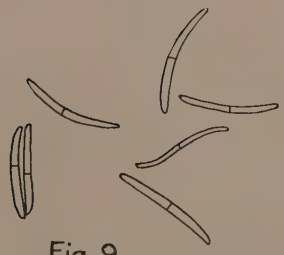

Fig. 9

Explanation of Figures

Fig 1. *Diatrypella borassi*
- a. Stroma with perithecia ×20
- b. Asci with Ascospores ×350
- c. Ascospores ×800

Fig. 2. *Glomerella alstoniae*
- a. Stroma with perithecia ×155
- b. Asci with Ascospores ×300
- c. Ascospores × 300
- d. Germinating ascospores ×280

Fig. 3. *Rosellinia erianthi*
- a. Perithecium showing Asci & Ascospores × 20 (Semi-diagramatic)
- b. Ascospores ×350

Fig. 4.
- a. T. S. leaf through uredo pustule of *Hyalopsora orientalis* ×72
- b. Uredospores of *Hyalopsora orientalis* ×350
- c. Conidia of *Marsonina zanthoxyli* ×280
- d. Conidia of *Attractium indica* ×280
- e. Spores of *Arthrobotryum velutinum* ×300

Fig. 5.
- a. Immature fructification (Natural size).
- b. Mature fructification (Natural size).
- c. Spores ×300

Fig. 6. *Phoma sirodesmii*
- a. Conidia ×300
- b. Pycnidia ×300
- c. Pycnospores ×300

Fig. 7. Infected leaf of *Grewia asiatica* with *Phyllosticta grewiae*

Fig. 8. *Phyllosticta heterophragmae*
Section through the pycnidium ×300

Fig. 9. Pycnospores of *Septoria adhatodae* ×540

PHYTOPATHOLOGICAL NOTES

New Physiologic Race of Melampsora lini *(Ehrenb.) Lev., in India*—R.S. Vasudeva, C. L. Sethi and V. C. Lele. Only four races of *Melampsora lini* (Ehrenb.) Lev., the causal organism of linseed rust have so far been reported in this country. Since these races are apparently different than those recorded elsewhere, they have been provisionally designated as Races I_1, I_2, I_3 and I_4 (Lele-1952).*

During the study of collections for the occurrence of physiologic races one sample from 1952-53 crop from the Punjab showed reactions markedly different from those of the known races, particularly on *Argentina* (C. I. 705-1) and *Koto*. In the case of the former variety, which is either immune or highly resistant to all the previously recorded races, it produces semi-resistant type of reactions. Similarly on the flax variety *Koto* this race produced small resistant type of pustules though it is immune to all the previously reported four races.

The new race has now been designated as race I_5. For the sake of comparison the reactions of all the races of linseed rust so far isolated in this country are set out in the Table—Division of Mycology & Plant Pathology, Indian Agricultural Research Institute, New Delhi-12.

*Lele, V. C. (1952) Study of physiologic races in Linseed Rust *Melampsora lini* (Pers.) Lev. Indian Phytopathology, **5**: 121-127.

TABLE

Reactions of all the five races of "Melampsora lini" (Ehrenb.) Lev. on 16 Main and 5 Auxilliary differentials, so far met with in India.

Race	Locality & Host from which races were originally picked up	MAIN DIFFERENTIALS															AUXILLIARY DIFFERENTIALS.				
		Buda	Williston Golden	Williston Brown	Akmolinsk	J. W. S.	Pale Blue Crimped	Kenya	Abyssinian	Morye	Ottawa C I. 770-B	Bombay	New Land	Bolly Golden	Italia Roma	Leona	T. P. Blue	Argentine C.I. 705-1	Bison	Victory	Koto	Ghawa
I_1	SOHAWRA : Local-Stock.	i	R	R—	SR or S	R or SR	i	i	i	i	i	S	i	i	i	i or R	i	i or R	i	1	i	i
I_2	SABOUR : Local-Stock	i or R+	R	R—	SR or S—	R	i	i	i	i	i	S	i	i	i	i or R+	i	i or R+	i or S—	i	i	i
I_3	LUDHIANA : Hybrid C. 4-2 Buda (R)	i or R+	SR	R—	SR or S	R or SR	i	i	i	i or R+	i	S	i	i	i or R+	i	i	i or R	i	i	i	1
I_4	JULLUNDUR : Local-Stock	SR or S	i or R+	R—	SR or S	R or SR	i	i	i	i	i	S	i	i	i	i or R+	i	i or R	i	1	i	i
I_5	GURDASPUR : Hybrid 54G Argentine (SR)	R— or SR	R	R—	R— or SR	R—	i	i	i	i	i	S	i	i	i	i or R—	i	SR	R— or SR	i	R or R—	i

i=Immune, R=Resistant, SR=Semi resistant, S=Susceptible

Note : —Minus sign indicates somewhat less resistance or susceptibility & plus sign somewhat higher resistance or susceptibility than shown by letters designating normal host reactions.

Factors Affecting Variability in Cereal Rust Reactions III. Variability due to age of the host*—T. N. Shukla. In 1950 race 15B, a biotype of race 15 of *Puccinia graminis tritici*, became widespread and prevalent causing considerable damage to the hitherto resistant wheats, especially to the durums, at some places in northwestern Minnesota and North Dakota in the U. S. A.

Tests in the field and greenhouse had indicated that all previously-grown, commercially desirable wheat varieties in the United States were susceptible, at least to some extent, to race 15B. Breeding for resistance to this race, therefore, already was in progress prior to 1950, because there was precedence for supposing that 15B, like other races before, might become more abundant and widespread.

Several varieties were used as resistant parents in breeding against race 15B. It was known that the resistance of some varieties varied with temperature, and possibly other factors, while that of other varieties remained more stable. Experiments dealing with the variability in rust reactions caused by temperature and light have been already reported by the author (Shukla, 1953, 1954). In the investigations under report the writer studied the susceptibility of three varieties to race 15B of *P. graminis tritici* at different stages of the development of the host.

Experimental—In the first experiment six-and 36-day-old plants of Lee, Stewart, and Kenya 58 wheats grown at 75°F. were inoculated with race 15B. The 36-day-old plants of Lee wheat were heading but those of Stewart and K58 were still in the boot stage. In the second experiment six-and 60-day-old plants of the same varieties grown at 75°F. were inoculated with the same race. The 60-day-old plants of Stewart and K58 had headed and the plants of Lee were in the mid-dough stage. After 48 hours in the moist chamber the plants were transferred to a greenhouse. In the first experiment (September 1951) the temperature fluctuated from 75° to 90°F. or higher during the day, and from 60° to 75°F. at night. The maximum light intensity for most of the days was 2500 to 6000 foot candles. In the second experiment (October 1951) the daytime temperatures fluctuated from 75° to 90°F. or higher. For the first three days the night temperatures ranged from 55° to 75°F., after that the night temperatures ranged from 73° to 80°F. Maximum light intensity for most of the time varied from 2000 to 3400 foot candles. In both the experiments rust notes were taken 20 days after inoculation.

Lee and Stewart plants of all ages were very susceptible to race 15B (Table 1). Six-day-old seedlings of K58 were abundantly infected with an infection type of 3 cn and had a general leaf tip necrosis. But on the 36-and 60-day-old plants the prevalence of rust was only moderate and infection type 1 to 1++ and no general necrosis of the leaf tip was noticed. The lower infection type on the older plants of K58 seems to be due to increased age.

* This work was carried at the University of Minnesota, U. S. A. under the guidance of Drs. E. C. Stakman and Helen Hart. Sincere thanks are due to them.

TABLE 1

Effect of age of the plant on the development of P. graminis tritici *race 15B on three wheat varieties in the greenhouse at 75°—90°F. during day and 65°—75°F. or more during night.*

Time of experiment, stage and age of the plant at the time of inoculation	Infection type 20 days after inoculation[a]		
	Lee	Stewart	K58
Sept. 4-24, 1951			
Seedling — 6 days	4 to 4+	4+ to 4++	3=cn[b]
Boot[c] —36 days	4 to 4+	4+ to 4++	1 to 1++
October 1-20, 1951			
Seedling — 6 days	4 to 4+	4+ to 4++	3=cn[b]
Headed[d] — 60 days	4 to 4+	4+ to 4++	1 to 1++

a The prevalence of rust was abundant in all cases expect 36-and 60 days old plants of K58 where it was only moderate.
b General leaf tip necrosis also was produced.
c Lee was heading.
d Plants of Lee wheat were in mid-dough stage.

CONCLUSIONS

The effect of age of the host on the development of rust varied with the variety. The susceptibility of Lee and Stewart wheats to race 15B of *Puccinia graminis tritici* was not affected by the age of the host. However, the older plants of Kenya 58 were resistant to 15B. —Laboratory of the Plant Pathologist to Government, U. P , Kanpur.

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Mutation in Puccinia graminis tritici *(Pers.) Eriks. & Henn., Physiologic Race 194—L. M. Joshi and Dulari Kak.* Mutation involving changes either in pathogenicity or colour or both are not frequent in cereal rusts and have been recorded by Newton and Johnson (1927), Waterhouse (1929), Stakman *et al* (1930), D'Oliveira (1929) etc. Colour mutation in race 15-C of Black Rust *Puccinia graminis tritici* (Pers.) Eriks.& Henn. was recorded by Misra and Lele

(1955). The case discussed in this paper however deals with two simultaneous mutations brought about in a single pustule of a pure race. In February 1954 a pustule of race 194 of black rust showed slightly yellow coloured portions at the periphery and isolations from this portion indicated that spores had undergone a change in certain morphological characters particularly in size and colour. The culture was maintained as such for eight months during which it passed through 11 successive generations without reversion to the original form. In October i.e., after about ten months, single spore cultures were established and it was then discovered that there were actually two mutants. Both the mutants have completed six generations and have been found to be consistant in their characters.

Parent race 194 of *Puccinia graminis tritici* was first isolated in 1945 from a wheat collection from Betul in Madhya Bharat and a single spore culture of the race was established at the Rust Research Sub-Station, Simla. When the mutations were first observed the race had completed 115 generations without any visible change in the morphological characters, or loss in virulence. The colour of the uredospores of the parent race in early stages is Amber Brown but with age gradually changes to Sudan Brown. The incubation period at an average range of temperature of 15° to 25°C is 8 days. The size of the spores varies between 24 to 41μ × 12 to 21μ and average length and breadth of the uredospores (based on the average of 100 spores) is 31μ and 18μ respectively. Characteristics of mutants are compared with the parent race in the following table :—

Culture	Colour of uredospores		Size of uredospores		Incubation period (15°-25°C)
	Age 18 days	Age 28 days	Range	Average	
Parent Race 194	Amber Brown to Sudan Brown	Sudan Brown	24 to 41 μ × 12 to 21 μ	31 μ × 18 μ	8 days
Mutant-1	Light Orange Yellow to Deep Chrome	Deep Chrome	18 to 31.5 μ × 15 to 19 μ	25 μ × 17 μ	9 days
Mutant-2	Raw Sienna to Mars Yellow	Mars Yellow	21 to 24.5 μ × 15 to 24 μ	28 μ × 18 μ	9 days

The mutants did not show any significant departure from the parent race in pathogenicity on the differential hosts and a few other wheat varieties except that there was slight difference in the incubation period. The mutants however, were apparently weaker than the parent race as the sporulation in both was comparatively less. Slight differences in the germination of uredospores were also observed. At the room temperature the uredospores of all the 3 cultures germinated in about 95 minutes. There was no appreciable difference in the early stages of germination but after about 2 hours the rate of growth of the parent race appeared to be much faster than that of Mutant—1. There was however, little difference in the germination rate of mutant—2.

Authors are deeply indebted to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his guidance during the course of these studies and in preparing the manuscript.—Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

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Rootrot of Chilli and its control. T. S. Ramakrishnan and A. P. Sarojini Damodaran. In and around Coimbatore, rootrot of chillies (*Capsicum annum*) has been prevalent in recent years. The diseased plants which could be easily spotted by the sudden wilting of the leaves even after irrigation, do not recover but eventually die. The incidence of the disease was usually observed when the plants had commenced to bear. The roots of the affected plants were discoloured and rotten. In some instances the collar region was also involved, with white strands of fungus mycelium growing over it.

Sclerotium rolfsii was isolated from the roots of the affected plants. Inoculations with this isolate produced hundred per cent infection on chilli plants while the controls remained healthy. The isolate was also pathogenic to groundnut, ginger and *Zinnia*.

The infection is soil-borne. The fungus produces large numbers of sclerotia which get mixed with the soil and remain viable for a long time. This renders the control of diseases caused by this fungus rather difficult. However the efficacy of three fungicides commonly used for the treatment of soils was tested for the control of this disease and the results are described.

Pure cultures of the fungus grown on sand-oats medium were mixed with sterilised soil in pots. Bordeaux mixture (5 : 5 : 50), Cheshunt compound (half an ounce in one gallon of water) or ceresan* wet (0.1 per cent solution) were added to the soil in the pots at the rate of half gallon per square yard of surface. Healthy chilli plants were transplanted in the pots, 24 hours later Two sets of controls were maintained. In one set the soil was inoculated but no fungicide was added. In the other set neither the inoculum nor the fungicide were added. In the course of a fortnight it was observed that all the plants in the first set of control, 90 per cent of the plants in the pots treated with Cheshunt compound and 88 per cent of the plants in the pots treated with Bordeaux mixture had been killed by the disease. But in the pots treated with ceresan and in the second set of control there was no incidence of the disease. *Sclerotium rolfsii* was isolated from the diseased plants in all cases. The experiment was repeated in field plots also and again it was found that ceresan was highly efficacious in completely preventing infection.

Based on the results of these experiments ceresan (wet) was recommended for use by the farmers in their fields. The rate of application recommended was half a pint of 0.1 per cent solution per plant to be used in the soil round the base. The treatment prevented the spread of the disease from the infected to the neighbouring plants. It had no effect however in curing the plants already infected. In some instances it may be necessary to repeat the treatment two to three weeks later.

Pots treated with Bordeaux mixture (1) and ceresan (2) and control (3) first set

*Ethyl mercury chloride.

The occurrence of Epicoccum purpurascens *Ehrenb. on Gladiolus and Pea.* H. K. Saksena. The genus *Epicoccum* Link, among the dark spored members of the family Tuberculariaceae, is characterised by dark globose sporodochia with spherical conidia. The conidia are often said to be one-celled but may become reticulate when the spores appear many celled. Some of the species of this genus are reported to be parasitic and some seem to be saprophytes. This paper gives a short account of *Epicoccum purpurascens* Ehrenb., first collected by the author on the leaves of gladiolus in Australia and subsequently, on pea leaves in India. As far as it could be ascertained, this fungus has not been reported on these two host genera and is also a new record for Australia and India. Robak (1932) has reported *E. purpurascens* on wood pulp from Norway showing slight attack on fibres and Hopkins (1932) records it from Southern Rhodesia where it caused cotton boll rot. It is also reported on pine and spruce of U. S. S. R. and from citrus in South Africa (R. A. M. 15 : 68).

Epicoccum purpurascens Ehrenb. Saccardo, *Syll. Fung.* 4 : 736, 1886.

1. On living leaves of *Gladiolus* sp. (cultivated) ; Uraidla, South Australia, 27.2.1952.

Sporodochia black, amphigenous, punctiform, globose to subglobose, crowded together in dense groups or rarely scattered irregularly and measuring 110–150μ in diameter, are placed upon rectangular to spreading pale yellow leaf spots. Mycelium of brown hyphae, 3.5-6μ thick, is irregularly branched forming a loose net work of wavy meshes.

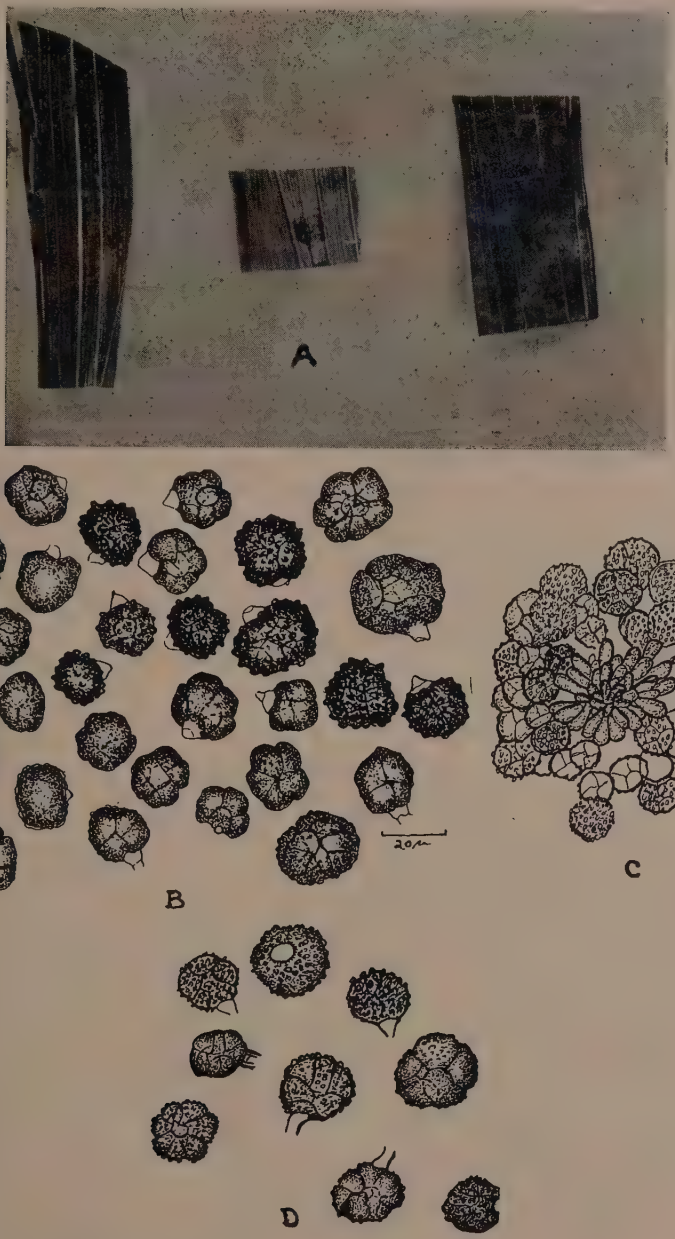
Conidiophores radiating from the sporodochia are simple and stout, 3-7μ long and bear mostly globose to sub globose, at first yellow at length brown, conidia measuring 14-24μ in diameter. The conidia are reticulate, each areola representing a cell, distinctly verrucose and consist of 4-10 cells with a hyaline to yellow pedicel tapering towards the base.

The specimen has been deposited in the Division of Plant Pathology, Waite Agricultural Research Institute. Adelaide.

2. On living leaves of *Pisum sativum* L. Beli, Allahabad, 28.3.1953; experimental plots, Govt. Agricultural College, Kanpur, 2.4.1955, H. K. Saksena.

On this host, the sporodochia mostly hypophyllous or amphigenous, measuring 90-150μ in diameter are placed upon brown elongated leaf spots bordered by a yellow zone. The conidia measure 13-22μ in diameter. The description of the other characters of the fungus is similar to that provided above and therefore it is identified as *E. purpurascens* Ehrenb.

On some leaves, colonies of *Alternaria* were found growing in close proximity to the sporodochial groups of *E. purpurascens* with their long conidial chains ramifying in between the sporodochia. This may then mask the otherwise distinct sporodochial formations.



Epicoccum purpurascens Ehrenb.

A. Sporodochia on the leaves of *Gladiolus* sp.
B. Mature conidia from A.
C. Young sporodochium from pea leaf showing the radial arrangement of conidiophores and young conidia
D. Conidia from pea.

The specimen is kept in the Botanical herbarium, Govt. Agricultural College, Kanpur.

Sincere thanks are due to Dr. C.G. Hansford, Head, Department of Plant Pathology, Waite Agricultural Research Institute, Adelaide for his help in the matter of specific identification of the fungus.—Plant Pathological Laboratory, Govt. Agricultural College, Kanpur.

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Robak, H (1932) [Investigations regarding fungi on Norwegian ground wood pulp and fungal infection at wood pulp mills.] *Nyt. Magazin for Naturvidens Kaberne*, B. Lxxi : 185 (Abst. in R. A. M. 12 : 69)

Studies in Aquatic Phycomycetes, II.—R. C. Lacy. The following nine fungi were collected and studied by the author in the course of his work at different places. This note is meant to supplement the record on Indian fungi, as these, as far as the author knows, are being reported for the first time from places noted against their names.

The fungi listed here were carefully studied and their characters compared with type descriptions for the purpose of their identification.

1. *Olpidium entophytum* (Braun) Rabenhorst, as in Sparrow, Aquatic Phycomycetes, pp. 88-89, 1943.

Parasitic on filament of *Spirogyra* sp., growing in a paddy field full of water, Dinapore, Patna (Bihar), 15-1-1950, R. C. Lacy.

2. *Rhizophydium minutum* Atkinson, in Bot., Gaz. 48 : 328, 1909.

Parasitic on filaments of *Spirogyra* sp., growing in a paddy field full of water, Dinapore, Patna (Bihar), 15-1-1950, R. C. Lacy.

3. *Blastocladia sparrowii* Indoh, as in Sparrow, Aquatic Phycomycetes, p. 433, 1943.

Saprophytic on a dead rotting turnip in a ditch of stagnant water, Machhwatoli, Patna, 25-2-1950, R. C. Lacy.

4. *Blastocladia rostrata* Minden, as in Sparrow, Aquatic Phycomycetes, p. 439, 1943.

Saprophytic on a dead rotting turnip in a ditch of stagnant water, Machhwatoli, Patna, 25-2-1950, R. C. Lacy.

5. *Blastocladia ramosa* Thaxter, as in Sparrow, Aquatic Phycomycetes, p. 440, 1943.

Saprophytic on a dead rotting turnip in a ditch of stagnant water, associated with *B. sparrowii* and *B. rostrata*. Machhwatoli, Patna. 25-2-1950, R. C. Lacy.

6. *Gonapodya prolifera* (Cornu) Fischer, as in Sparrow, Aquatic Phycomycetes. pp. 472-474, 1943.

Saprophytic on rotting apple pieces, placed as baits in foul wayside drain, Jalla, Patna, 24-4-1950, R. C. Lacy.

7. *Gonapodya polymorpha* Thaxter, as in Sparrow, Aquatic Phycomycetes, pp. 474-475, 1943.

Saprophytic on rotting apple pieces, placed as baits in foul wayside drain, Jalla, Patna, 28-4-1950, R. C. Lacy.

8. *Myzocytium proliferum* Schenk, as in Sparrow, Aquatic Phycomycetes, pp. 649-652, 1943.

Parasitic on *Spirogyra* sp., Jalla, Gulzarbagh, Patna, 25-5-1950 and Mac-Pherson Lake, Allahabad, 20-12-1952, R. C. Lacy.

9. *Dictyuchus monosporus* Leitgeb, in Couch, Jour. Elisha Mitchell Sc. Soc. 40 : 116, 1924.

Saprophytic on a piece of dead twig in a pond, Bahadurpur, Patna, 10-2-1950. R.C. Lacy.—Department of Botany, Ewing Christian College, Allahabad-3.

The Occurrence of Sclerospora philippinensis *Weston on "Kans Grass" (Saccharum spontaneum L.) in India*—B. L. Chona and D. Suryanarayana

In July, 1955, the writers observed yellowing of leaves in certain clumps of *Saccharum spontaneum* commonly known as '*Kans*' growing along the sides of an irrigation channel at the Indian Agricultural Research Institute, New Delhi. The affected leaves showed a downy white growth mostly on the under-surface. Microscopic examination revealed that it was the sporangial stage of the Peronosporaceous genus *Sclerospora*. The sporangiophores are stout with short branches at the top, emerging from the stomata and measure 144-325μ in length. The sporangiophore has a well-developed basal cell and is branched at the top having sporangia on the ultimate branches Sporangia are elongate-ovoid, elongate-ellipsoid or rotund-cyclindrical and measure 32-44.8$\times$16-22.5μ and germinate by means of germ tubes. Oospore stage is not observed. The mycelium of the fungus was found to be present in all the parts of the plant, thus indicating systemic infection.

From the foregoing, the pathogen is identified as *Sclerospora philippinensis* Weston. It is the first time that this fungus is recorded on this host in India.

Fig. 1—A young sporangiophore having sporangia still attached and a basal cell.

Fig. 2—A mature sporangiophore with all the sporangia detached.

Fig. 3—Upper part of the sporangiophore showing dichotomous branching.

Fig. 4—Sporangia.

Figs. 5 & 6—Sporangia germinating by germ tubes. In Fig. 6 the germ tube is branched.

(All the figures are magnified 325 times)

The occurrence of *Sclerospora philippinensis* on *Saccharum spontaneum*, which is a perennial wild grass throughout the warmer parts of India, indicates the possibility of its serving as a collateral host for the downy mildew of maize, which would explain the annual recurrence of the mildew despite the absence of the oospore stage of the fungus.

We are grateful to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi, for providing necessary facilities, helpful criticism and encouragement.—Division of Mycology and Plant, Pathology, I.A.R.I., New Delhi.

ERRATA TO VOL. VIII, No. 1, 1955

1. Page 18, foot-note to Table 3: Active ingredient of AGROSAN GN is phenyl mercury acetate, ethyl mercury chloride mixture, and not tolylmercury acetate.
2. Page 45 line 13 : in place of "p. 240" read "p. 48".
3. Page 53, second line of last para : delete '(vide Fig. 3)'.
4. Page 55 : delete Fig. 3 which was inserted due to editorial mistake.

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